An Erythristic Morph of Red-backed Salamander (*Plethodon cinereus*) Collected in Virginia

Trevor L. Chapman[1], Brian G. Gall[2], Kari L. Spivey

[1] East Tennessee State University, Johnson City, TN 37604
[2] Hanover College, Hanover, IN 47243

ABSTRACT

Eight different color polymorphisms of the Red-backed Salamander (*Plethodon cinereus*) have been described throughout its range. Among them, the erythristic phenotype is a mimetic morph with coloration patterns similar to that of toxic Eastern Newt efts (*Notophthalmus viridescens*). We describe an erythristic morph of *P. cinereus* collected at Mountain Lake Biological Station, Virginia. To our knowledge, there are no prior published records of this morph in Virginia.

Key words: *Plethodon cinereus*, *Notophthalmus viridescens*, erythristic

The Red-Backed Salamander (*Plethodon cinereus*) is widely distributed throughout eastern North America, with a range reaching as far south as North Carolina and north into Maine and parts of Canada (Mitchell and Gibbons 2010). These terrestrial plethodontids most commonly occur in populations with two distinct phenotypic morphs: red-backed and lead-backed phases. The body of both phases is characterized as dark gray to black with white flecking, and the ventral surface is black with very small white spots. The red-backed phase is differentiated from the lead-backed by a broad, straight-edged stripe down the dorsal surface from snout to tail. This stripe can occur in orange, yellow, or red coloration. Although these two morphs are exceedingly predominant in Red-backed Salamander populations, a total of eight color polymorphisms has been described throughout its range (Moore and Ouellet 2014). The other six polymorphisms are albino, amelanistic, erythristic, iridisctic, leucistic, and melanistic. The six rare morphs are reported mostly in the mid-north part of the species range.

The erythristic morph is characterized by a reddish-orange body lacking a defined stripe, with black mottling in varying degrees along the sides and dorsum. The coloration is similar to that of Eastern Newt efts (*Notophthalmus viridescens*), which contains small amounts of tetrototoxin, a neurotoxin secreted from the skin (Brodie 1968). Although Lotter (1975) found no correlation between occurrence of erythristic morphs and number of efts in various localities in the northeastern United States, it has been hypothesized that the polymorphism is a form of Batesian mimicry (Lotter and Scott 1977). Among the rare color patterns, only this morph has

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been demonstrated to be an adaptive color polymorphism. This notion was supported in a study by Brodie and Brodie (1980) in which *N. viridescens* efts, Allegheny Mountain Dusky Salamanders (*Desmognathus ochrophaeus*), and both red-backed and erythristic morphs were exposed to avian predators. Survival in eft newts was the highest, but survival in the erythristic morphs was significantly higher than that of Red-backed morphs and *D. ochrophaeus*. Currently, erythristic morphs are known from 10 states, the two most southern being Ohio and Maryland.

On 15 May 2017 we discovered an erythristic morph of a red-backed salamander at Mountain Lake Biological Station, Virginia. It was under a small decomposing log with mostly dry substrate on a north-facing slope approximately 50 m west of Spring Trail. Other terrestrial salamander species found within 100 m included Eastern Newt efts (*Notophthalmus viridescens*), Northern slimy salamanders (*Plethodon glutinosus*), and Gray-cheeked salamanders (*Plethodon montanus*), as well as both red-backed and lead-backed morphs of *P. cinereus*.

The erythristic morph was an adult male with a total length of 65 mm. The dorsum was solid red-orange with some black mottling (Figure 1). The erythristic coloration also covered the head and extended down the sides, becoming more broken towards the venter. The proximal portions of all four limbs were the same color as the dorsum until the first joint, after which they were mostly black. The red-orange coloration extended down the dorsal surface of the tail, with the sides and ventral surface of the tail being mostly black.

Figure 1. *Plethodon cinereus* from Mountain Lake Biological Station, Virginia, with erythristic phenotype.

To our knowledge, this is the first time that an erythristic morph of a red-backed salamander has been recorded in Virginia. Furthermore, this morph has never been reported south of Maryland (Moore and Ouellet 2014). Mitchell et al. (2003) described a red-backed salamander with an unusual color phenotype in northern Virginia, but the authors do not describe it as erythristic in the report. The authors also cited a personal communication with E. Lanham, who claimed to have observed an erythristic morph on Mountain Lake property in 2002. However, the salamander was not described or photographed. While the morph is most common in cooler climates of glaciated areas in the northeastern United States, Pauley et al. (2001) reported a population in Pennsylvania consisting of 47.4% erythristic morphs. The authors speculated that the high elevation and low air temperatures at the location might explain the occurrence and high frequency. Given that the elevation at Mountain Lake Biological Station exceeds 1,200 m, it is a likely habitat in Virginia to find a morph that is more typical of a northern location.

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Population Dynamics of the Cotton Rat in Southeastern Virginia

Robert K. Rose[1] and Heather Green Salamone[2]

[1]Department of Biological Sciences, Old Dominion University, Norfolk, VA 23529-0266
Corresponding author: brose@odu.edu
[2]15 Fillmore Avenue, Danbury, CT 06811

ABSTRACT

We used monthly live trapping for 2.5 years to evaluate the life-history features of the most common small mammal, *Sigmodon hispidus* (hispid cotton rat), in an old field at its northern limit of distribution on the Atlantic Coast. Peak densities, achieved in late autumn or early winter, were among the highest recorded for the species and were more typical of geographically marginal populations rather than of central ones. Unlike some other marginal populations, hispid cotton rats in southeastern Virginia did not lose significant body mass over the winter (when few juveniles were present) and survival in winter was not significantly different from that of other seasons, perhaps due to the moderating effects on winter temperatures of the nearby Atlantic Ocean and Chesapeake Bay. Our study provides support for the presence of spring and autumn cohorts, with long-lived animals being drawn almost entirely from the latter.

INTRODUCTION

Sigmodon hispidus Say and Ord (hispid cotton rat), a 100-g herbivorous rodent, is the sole member of a tropical genus broadly distributed across the southern US, for which the northward expansion of its distributional range into the central states and along the Atlantic Coast is well known (Cameron and McClure, 1988). Since the first Virginia report (Patton, 1941), it has spread across southern Virginia and into the Piedmont (Pagels and Moncrief, 2015), and likely is spreading northward wherever its movements are not impeded by large rivers or the Chesapeake Bay. Although much studied across its distribution, capture-mark-recapture (CMR) studies that provide the most useful information on demographic features are limited to those in Florida (Layne, 1974; Stafford and Stout, 1983), Texas (Cameron and colleagues), Oklahoma (Goertz, 1964; McMurray et al., 1994; Schetter et al., 1998), eastern Kansas (Slade and colleagues), and western Kansas (Fleharty et al., 1972); this report provides demographic information for a marginal population in coastal Virginia.

Unlike many temperate rodents, the population biology of the hispid cotton rat (hereafter, cotton rat) is highly variable across its distribution, with some populations showing high breeding activity in spring and autumn, and others only in autumn (Cameron and McClure, 1988). Peak densities in late autumn, the usual pattern for north temperate rodents, are reported

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in some populations, but a population in Florida had a peak in August and still others had December or January peaks. Numerous studies report extirpations or near extirpations of local populations in Oklahoma, Kansas, and Tennessee (Dunaway and Kaye, 1961; Goertz, 1964; Sauer, 1985; Langley and Shure, 1988; Clark et al., 2003), an indication of a lack of adaptation to severe cold. Further, cotton rats in these northern regions often lose body mass over the winter (Dunaway and Kaye, 1961,1964; Goertz, 1965; Chipman, 1966; Slade et al., 1984; Schetter et al., 1998) but adults in other populations (e.g., Georgia, Virginia) maintain or gain body mass during winter (Bergstrom and Rose, 2004; Green and Rose, 2009). Sex ratios often deviate from unity, and in unpredictable ways.

The objective of our study was to provide demographic details for a population of cotton rats from southeastern Virginia, based on monthly live trapping across a 2.5-year period that included 3 winters, including patterns of density, sex ratio, body mass, and residency, plus length of reproductive seasons, proportions of transients, and lifespan (longevity).

MATERIALS AND METHODS

Description of field site – Our study site was an old field owned The Nature Conservancy located in southern Chesapeake (37º50'N, 76º20'W), Virginia. When we began the field study in December 2002, the land had been withdrawn from agricultural production for 2 years and was dominated by chest-high little bluestem (*Schizachyrium scoparium*) with other associated grasses, mostly panic grasses (*Panicum* spp.), and with short volunteer trees, mostly loblolly pine (*Pinus taeda*). The 11.5-ha field was bordered on the north and east by mature hardwood forest, on the west by a road and mature pines beyond that, and on the south by a freshwater marsh. The study grid was bisected by a meter-wide and 0.5 m-deep drainage ditch that filled during the winter months, typical of this region of high water table. The site lies about 4 km east of the Great Dismal Swamp National Wildlife Refuge.

Field methods – We trapped for 3 days each month from December 2002 through July 2005, except for June 2003 when extreme predator disturbance required closing the traps. Our study grid was 8 by 8 with 12.5-m intervals, producing a grid with an effective trapping area of 1 ha (Stickel, 1954). At each grid coordinate we placed 2 Fitch live traps (Rose, 1994) baited with a mixture of wild bird seed and sunflower seeds, with fiberfill added in winter for insulation. We set traps in the late afternoon, usually during the new moon phase, and checked them early for the next 3 mornings. From April through October, we locked the traps open in the morning and reset them again just before sundown to prevent heat-related mortality in the traps. Mortality for rodents was nearly zero.

Each small mammal was given a right ear tag with unique number, which, if lost, was replaced with a tag in the left ear, and the animal was synonymized to avoid inflating estimates of density. We recorded information on reproductive condition of males using abdominal testes (not fertile) versus descended testes (fertile). We evaluated 3 reproductive features in females: not perforate or perforate vaginal orifice, small-medium-large nipple size, and closed-slightly open-open pubic symphyses; the latter conditions in each category are associated with reproduction. We used a Pesola™ pencil-scale to determine body weight (g), then released the animal at the coordinate of capture. We collected the same information for recaptured animals,

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although within a month only next coordinate was recorded. Our goal was to mark all animals and follow events of their lives. We defined juveniles as those < 50 g, sub-adults as 51–100 g, and adults as those > 100 g. We used a 50-g criterion as the upper limit for juveniles because our earlier necropsy studies had revealed fertile males and pregnant females weighing less than 60 g in populations from southeastern Virginia (Rose and Mitchell, 1990; Bergstrom and Rose, 2004). We designated 3 residency classes: transients (seen only in 1 month), visitors (seen in 2 months), and residents ($\geq$ 3 months).

We conducted our study before our university IUCAC required approval for field studies of wild mammals. Our methods followed the guidelines for the use of mammals in research, as outlined by the American Society of Mammalogists, the latest version of which is Sikes et al. (2016).

Statistical analyses – We used SPSS version 12.0 (2003; SPSS, Inc., Chicago, Illinois) for analysis. Mean masses for both sexes were calculated for month and season. We defined winter as December-February, spring as March-May, summer as June-August, and autumn as September-November.

We used Chi-square tests to determine deviations from unity in sex ratios and to test for differences among the three residency classes. We determined density using the JOLLY (Hines, 1996) software package, which uses the Jolly-Seber model, and also calculated density by hand using minimum number known to be alive (MNA: Krebs, 1966), which is calculated by adding the number of individuals captured during month t to those tagged animals but not captured in month t but known to have survived to month $t + 1$ or beyond. Importantly, we trapped beyond February 2005 (the end of our period of analysis) because the population estimators require the numbers of animals captured months later to enable accurate calculations. Thus, we monitored the decline of the population after February 2005 but present only anecdotal or qualitative information from the spring-early summer 2005 period.

We used correlation analysis to examine the concordance of the 2 methods of estimation and also used the density values produced by JOLLY to evaluate a possible correlation between the first and second year of the study. We used Student's *t*-tests to detect sexual dimorphism and two-sample *t*-tests to examine body mass differences between years. We analyzed mean mass using a model-I two-factor analysis of variance (ANOVA) to observe any potential significant differences between sexes and among months. We used REGWF multiple range tests to identify important variables for each ANOVA for which factors were significant (SPSS, 2003).

RESULTS

We tagged 864 different small mammals of 8 species in 9088 trapping nights, of which *Sigmodon hispidus* accounted for 513 (59.4%) individuals. (Table 1 shows the sum of the tagged animals caught each month and the number in parenthesis includes recaptures within a month.) Meadow voles (*Microtus pennsylvanicus*) and eastern harvest mice (*Reithrodontomys humulis*) were sub-dominant members of the community. The other mammals were much less common and sometimes their presence was brief (e.g., no house mice, *Mus musculus*, after May 2003) or

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intermittent (marsh rice rats, *Oryzomys palustris*, were present mostly in the cool months, often also the wettest ones).

Population density – Using the best-fit model (Jolly-B: $X^2 = 46.29$, $df = 33$, $P = 0.062$) for this population, density in late winter 2003 was estimated to be about 60 cotton rats/ha, but numbers dropped by half that spring before recovering to comparable density in late summer (Fig. 1). By the end of the autumnal breeding season, population density had exceeded 100/ha at the start of the second winter. The highest densities (124/ha) were achieved in January 2005, in part due to the breeding season extending into November in 2004. The decline in February 2005 continued into spring and summer and when we ended monthly trapping in July 2005, only a few cotton rats remained (we caught three in July). The Pearson's correlation between JOLLY and MNA values was highly significant ($r = 0.963$, $n = 25$, $P < 0.001$), validating the density estimate of either method.

Sex ratios – The sex ratio (248 males: 265 females) of all tagged cotton rats was not different from unity ($X^2 = 0.56$, $P > 0.50$). However, tagged females were trapped in more successive months than males, so when all tagged cotton rats caught at least once per month are considered, we captured significantly more females than males (652:538; $X^2 = 10.92$, $P < 0.001$). This ratio is unusual because in most small mammal studies, captures of males outnumber those of females by approximately 3:2.

Age distributions – Across the study, juveniles comprised 13 % of each sex. Thus, the population was dominated by sub-adult and adult cotton rats. During the first 2 winters, less than 5 % of cotton rats were juveniles and in the third winter no juvenile was present.

Patterns of residency – Our population showed a high proportion of residents: 72 percent (Table 2), and thus relatively low proportions of transients and visitors. For visitor and resident classes, some tagged animals were absent or not captured for 2 or 3 months and then returned to the trappable population (7% of males, $n = 17$, and 6% of females, $n = 15$). Female residents were significantly more numerous than male residents (543 versus 428: $X^2 = 13.62$, $P < 0.001$) but more male than female transients were recorded ($X^2 = 6.95$, $P < 0.05$).

Patterns of reproduction – Distinctive breeding and non-breeding seasons were evident based on external features of both sexes. In all 3 winters, neither males nor females showed signs of breeding, but by March males had descended testes and by April about two-thirds of females had perforate vaginae and enlarging nipples. Females gradually increased their level of reproduction from August through the end of the year. Testicular regression, begun in October, was completed by December. This population was characterized by few young being raised in spring or early summer but greatest recruitment occurring in autumn, and extending into January in the last year.

Patterns of body mass – For an assessment of body mass, we considered only those cotton rats $\geq$ 50 g in the analysis and also excluded obviously pregnant females. Because each tagged sub-adult and adult cotton rat likely has a different body mass each month, we included each weight at first capture of a month in calculations of body mass. The mean body mass for males was 99.51 ± SE 1.327 g ($n = 466$) and 92.23 ± SE 1.132 g ($n = 522$) for females. These means were significantly different ($t = 4.007$, $P = 0.001$). Further, males were significantly heavier in 2003

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(104.71 ± SE 2.002 g) than in 2004 (97.92 ± SE 2.218 g; $t = 1.931$, $P = 0.054$), but females had nearly identical mean masses in both years. Male residents were significantly heavier that male transients ($t = -2.655$, $P = 0.01$) but female residents and transients were of similar size.

A model-I two-factor ANOVA using sex and months as factors on log-transformed data confirmed the significant mass differences between the sexes ($F = 7.451$, $df = 1959$, $P = 0.006$) and also significant differences among months ($F = 4.97$, $df = 25,958$, $P = 0.001$). The sex-month interaction term was also significant ($F = 2.046$, $df = 22,958$, $P = 0.03$).

Despite no significant differences in mean mass for winter months, as demonstrated by the REGWF tests from the model-I two-factor ANOVA on monthly values, we observed slight, irregular variations in mean mass in all 3 winters (Fig. 2). The mean mass of females was stable in 2 winters but decreased slightly in the second winter. Males were more erratic, gaining body mass the first winter, losing some the second winter, and holding constant mass the third winter. Overall, masses of both males and females were nearly constant over the winter months.

Longevity – We examined the lifespans of the 12 males and 21 females recorded on the grid for 6 or more months; their mean longevity was similar, 7.2 and 7.5 months, respectively. Except for those tagged in December at the start of the study, most long-lived cotton rats were tagged at juvenile body masses, usually 20–40 g. One adult female, first caught in October 2003, was captured multiple times, had litters in May and October 2004, and likely was born in April or May 2003, making it about 20 months old when last caught in January 2005.

DISCUSSION

We observed peak densities in late autumn or early winter, with ~60/ha, 100/ha and 124/ha in the 3 years, with declining densities through winter but with lows not dipping below about 30/ha during the first 2 springs. However, at the end of the study, the pines had shaded out the grasses, greatly diminishing the monocot-rich diet of cotton rats (Walker and Rose, 2009) and causing some to rely on pine bark for part of their nutritional needs during the late winter and early spring of 2005 (Nadolny and Rose, 2015). In the last spring, the population did not recover, with numbers dwindling to (probably) below 10/ha in summer. Although the month of lowest numbers was May in the first spring and February in the second spring, there was no hint of even a minor peak of breeding in spring 2005 in our population (not shown in Fig. 1 for the reasons explained in Statistical Analyses in Methods).

The densities we observed (100/ha and 124/ha) are among the highest reported in comparable CMR studies. Schetter et al. (1998), in eastern Oklahoma, recorded peak densities of 90 and 110/ha on 2 grids in year 1 and 55/ha on both grids in year 2; peaks of both years were in August. Wilson et al. (2006), also in eastern Oklahoma, also report highest densities (86/ha) in August, whereas in central Oklahoma peak density (112.5/ha) was achieved in October (Clark et al., 2003). In northern Georgia, highest densities (118/ha and 88/ha) were recorded in May in 2 old fields (Langley and Shure, 1988). In central Florida, peak density of 47/ha was achieved in December, a value that dropped to 10/ha the next month (Stafford and Stout, 1983). Other peak densities were 4.4/ha (Layne, 1974), also in central Florida, and 14/ha across 3 years of study in coastal prairie habitat in Texas (Cameron, 1977). In eastern Kansas, highest densities, reached in

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autumns of 9 years, were about 26–34/ha (Diffendorfer et al., 1995), whereas in a later year the highest density on a control grid was 39.5/ha (Doonan and Slade, 1995); their 2 food-supplemented grids achieved densities of 100–110/ha. In much drier western Kansas, the peak density was 24/ha (Fleharty et al., 1972). In brief, peak density for a region is highly variable, possibly dependent on primary production of the habitat, and is achieved in different seasons, sometimes in summer, more commonly in autumn, but even in winter, as we observed in January 2005. If there is a pattern in these several studies it is that marginal populations, living in more seasonal climates, tend to achieve higher peak densities than the more central populations in Texas and Florida. Furthermore, some studies (e.g., Langley and Shure, 1988) report that severe winter was followed by greatly reduced primary production and the slow recovery of a cotton rat population, further support for the importance of primary production and nutrition.

Tagged cotton rats had a 1:1 sex ratio in our study, but females outnumbered males when we counted the number of tagged animals caught at least once in a month. The most likely explanation for this result is that more females than males were residents and on average females had more captures per individual than males: both were true. In some months, captures of females were much more numerous than males, which is unusual. Male rodents usually have larger home ranges than females and therefore are viewed as being prone to more captures per individual than females. Further, we recorded significantly more transient males than females, an expected result.

Although most studies of cotton rats report sex ratios of unity (e.g., Layne, 1974; Cameron and McClure, 1988; and our study for all tagged animals), Schetter et al. (1998) observed sex ratios of 60–65% males in February and May in both high- and low-density populations in Oklahoma. Goertz (1965), also in Oklahoma, reported male-biased (60%) sex ratios during one May–September period. Joule and Cameron (1980) recorded fluctuating sex ratios, with values sometimes reaching 60% males and at other times 60% females, with no apparent seasonal pattern. During the breeding season, when females (but not males) seek patches of dicots in the Texas coastal prairie (Cameron and Spencer, 2008), even the habitat quality of a grid possibly affects sex ratio. Our population had months and even seasons when females were notably and even significantly more numerous than males; it was as if males had died or emigrated. For example, in autumn and winter 2003-2004 (Table 2), 95 males and 172 females were recorded, a huge departure from unity. Thus, unlike many species of small mammals, cotton rats seemingly have varying and perhaps fluctuating sex ratios, sometimes within the same population, a pattern awaiting an explanation.

We used a 50-g criterion as the upper limit for juveniles because our earlier necropsy studies had revealed fertile males and pregnant females weighing less than 60 g in populations from southeastern Virginia (Rose and Mitchell, 1990; Bergstrom and Rose, 2004). Our use of a 50-g criterion likely contributed to the low percentage of juveniles (13% for each sex) compared to other studies that used a 60-g criterion, such as Stafford and Stout (1983), who report 28% of males and 40% of females were juveniles in their populations in central Florida. In coastal Texas, Cameron and Kruchek (2005: Fig 4) show juveniles comprising 10–12% of the population in spring and summer but their virtual absence in other seasons. In contrast, Layne (1974), another who used a 50-g criterion, reported 25% juveniles in his population in Florida.

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The faster growth rates in northern populations than in more central ones likely contributes to the generally low proportion of juveniles in marginal populations.

Compared to some other studies, we believe our population had a high proportion of resident cotton rats (Table 2), probably due in part to unsuitable habitat in 3 directions from the grid and a brushy wetland on the fourth. We estimated that proportion by the numbers of resident males and females from each season in Table 2 as a percentage of the row totals. Across the study, about 72 percent of cotton rats were residents, using the criterion of captures in 3 or more months to define a resident. Other investigators have examined transiency patterns; Doonan and Slade (1995) found that 21% of cotton rats were transients on their control grid, and 28% were transients on their food-supplemented grid. Also in eastern Kansas, the percentages of transients ranged from less than 1 to 67 % per month, with the mean being 29 % (Diffendorfer et al., 1995). Among seasons, we observed many fewer transients in winter than in spring or autumn, suggesting that higher proportions of animals were relatively more place-bound in winter.

Cotton rats in southeastern Virginia breed from March through October (Rose and Mitchell, 1990; Bergstrom and Rose, 2004) with occasional extensions by females into November, as happened in 2004, leading to peak density 2 months later. In southeastern Virginia, breeding starts slowly in spring, with the first litters in April and May, but the greatest proportion of breeding females was observed in September and October. Some regional populations have similar patterns. For example, Stafford and Stout (1983) and Layne (1974) found some spring breeding but far greater reproductive activity in autumn in central Florida. In southern Florida, no breeding was detected during the December-May dry season (Smith and Vrieze, 1979). In Oklahoma, Goertz (1965) recorded pregnant females in every month except December and January and a mid-summer lull in breeding. Also in Oklahoma, McMurry et al. (1994) saw high levels (60–100%) of reproduction by females in summer and autumn, with lower levels of breeding in other seasons and none in one spring. In Arkansas, peak breeding from February-July was followed by less reproduction through November (Sealander and Walker, 1955). In eastern Tennessee, winter breeding was rare but breeding peaks were seen in June-July and September-October (Dunaway and Kaye, 1964). In coastal eastern Texas, breeding indices were substantially higher in spring and summer than in the other seasons (Cameron and Kruchek, 2005). The conclusion from reviewing these and other studies of reproduction is that cotton rats have adjusted their breeding seasons to their geographic locations, often starting earlier in the year in the south where a mid-summer lull is common, and with lower levels of breeding in spring by overwintered females than by spring-born females that likely have 1–3 litters in late summer and autumn. Whether our observation, that nearly 100 percent of cotton rats surviving the winter are born in late summer or autumn, is typical of other northern populations remains to be demonstrated.

In theory, high peak densities can be achieved, in part, by lowering the age at first reproduction, a feature observed in some marginal populations. For example, Goertz (1965) found 51 g and 53 g pregnant females in Oklahoma, similar to what has been observed in Virginia (Bergstrom and Rose, 2004; Rose and Mitchell, 1990). Because embryos are not visible until 10 days of pregnancy (Meyer and Meyer, 1944), a 55-g female likely is impregnated when weighing less than 45 g. Future studies may confirm that females from marginal populations begin breeding at lower body masses than those in more central populations.

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Virginia cotton rats are sexually dimorphic, with adult males averaging about 100 g and adult females about 8 g less (Bergstrom and Rose, 2004; Rose and Mitchell, 1990; our study). (Many early studies included the masses of juveniles in the analysis of body mass, thereby greatly lowering mean mass during months of recruitment of young into the population; the more useful comparisons are those that exclude juveniles, whether defined at 50 or 60 g.) In our study, mean seasonal body mass tended to be greatest in spring and lowest in winter; in some seasons the sexes had similar masses (Fig. 2). This pattern is reported for almost all other populations: sexual dimorphism is the rule but in some months the sexes have similar masses. A typical pattern of body mass is seen in coastal Texas (Cameron and Spencer, 1983), with rapid increases in body mass of both sexes in spring, followed by fluctuations in mass, and with mean body mass decreasing in winter.

Several investigators have reported reductions in adult body mass during winter: Chipman in Texas (1966), Goertz (1965) and Schetter et al. (1998) in Oklahoma, Slade et al. (1984) in Kansas, and Dunaway and Kaye (1961, 1964) in Tennessee. In central Florida, adults lose mass starting in October and winter weight loss is as pronounced as in more northerly populations (Layne, 1974). Further, no large adult survived from autumn to spring in Layne's study. Thus, even in central Florida, winter can exact a mass loss and mortality toll. In our population, both sexes nearly held their mean body masses over 3 winters.

The mean lifespans for long-lived males and females in our study are similar to those of previous studies, with several adults living more than 7 months and one female nearly 20 months. Determination of lifespan requires a long trapping history, which is difficult to achieve for a species as vagile as the hispid cotton rat. In his 14-month study in central Florida, Layne (1974) estimated mean residency time for both sexes and all age groups to be 2.9 months. Goertz (1964) reported that half of marked animals disappeared by the second month in Oklahoma and 98% were gone by the 6th month. This is contrasted by 16% of 180 cotton rats surviving for 6 months in Tennessee (Dunaway and Kaye, 1964). Much of gross mortality is attributable to continual movement (leading to emigration out of the study area) rather than to death, but Schnell (1964, 1968) used observational and experimental studies to document losses to a variety of both avian and terrestrial predators as well as natural deaths.

Some investigators (e.g., Goertz, 1965; Layne, 1974) report the loss of large adults as autumn grades into winter, also observed by Odum (1955), who inferred that cotton rat populations may have 2 seasonal cohorts, with few or no spring-born animals surviving the winter and with most long-lived animals being those born in autumn. In our study, no long-lived cotton rat was first caught and tagged from January to July; almost all were tagged between August and November, supporting this conjecture. We believe this pattern supports the argument of spring and fall cohorts in the life cycle of cotton rats.   In our population, the fall cohort was long-lived compared to the spring cohort, almost no member of which survived as long as 6 months. Of course, there are exceptions, such as the female in our study that lived through one winter and into the second, and Layne (1974) reporting 2 females tagged in the first month still being alive 14 months later.

In conclusion, our population of *Sigmodon hispidus* in southeastern Virginia was similar in some features to marginal populations in the central states by achieving high annual densities

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in late autumn (rather than in summer) and by having long-lived animals drawn mostly from the autumn cohort, but was different by (nearly) sustaining early winter body mass through the winter, probably due to the moderating effects on winter temperatures of the nearby Chesapeake Bay and Atlantic Ocean. In all, the hispid cotton rat, as a tropical rodent expanding into more temperate locations, is adapting to local conditions by adjusting its peak breeding periods to later in the growing season, perhaps to take advantage of greater plant productivity or nutritional quality of its primarily monocot foods.

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TABLE 1. Total numbers of small mammals caught in live traps from December 2002 through February 2005 in Chesapeake, southeastern Virginia. Each number is the sum of the different tagged animals caught each month and the number in parenthesis includes recaptures within a month. Sex could not be determined for the shrews and a few immature rodents.

Species	Males	Females	Unknown	Total
Sigmodon hispidus	538 (694)	652 (874)	0	1190 (1568)
Microtus pennsylvanicus	141 (181)	124 (153)	1	266 (335)
Reithrodontomys humulis	96 (104)	90 (115)	0	186 (219)
Mus musculus	43 (44)	19 (19)	1	63 (64)
Oryzomys palustris	20 (25)	14 (14)	1	35 (40)
Cryptotis parva	—	—	11	11 (11)
Blarina sp.	—	—	6	6 (6)
Microtus pinetorum	1 (2)	3 (3)	0	4 (5)
Total	839 (1050)	902 (1178)	20	1761 (2248)

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TABLE 2. Numbers of male and female *Sigmodon hispidus* that were transients, visitors, and residents as recorded in different seasons in Chesapeake, southeastern Virginia. Transients were observed in only one month, visitors in 2 consecutive months, and residents for at least 3 months on the grid. We estimated the percentage of residents in each season by dividing the numbers of residents from each season by the row totals; more than half of tagged cotton rats were residents in every season.

| Season	Transients		Visitors		Residents		% Residents
	Males	Females	Males	Females	Males	Females	
Winter 2002-2003	19	26	13	10	50	44	58
Spring 2003	9	9	5	4	47	45	77
Summer 2003	12	7	5	0	34	35	74
Autumn 2003	16	14	15	12	68	102	75
Winter 2003-2004	10	10	8	9	27	70	72
Spring 2004	20	3	3	1	18	34	66
Summer 2004	16	6	0	14	25	34	62
Autumn 2004	14	9	18	11	77	90	76
Winter 2004-2005	13	6	8	14	82	89	81
Total/Mean	129	90	75	75	428	543	72
Grand total	219		150		971		

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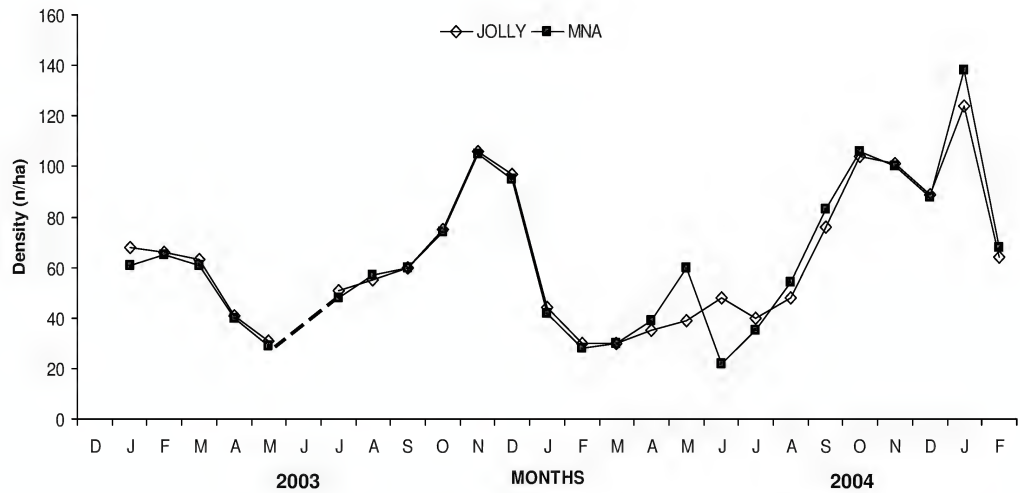
FIGURE 1. Monthly estimates of population density of *Sigmodon hispidus* from December 2002 to February 2005 in Chesapeake, southeastern Virginia. Density (*n*/ha) was determined by the software package JOLLY and by minimum number alive (MNA) methods. Dashed lines represent the gap in data collection for June 2003. Increases in density were observed in both autumns and slightly in the spring of 2004.
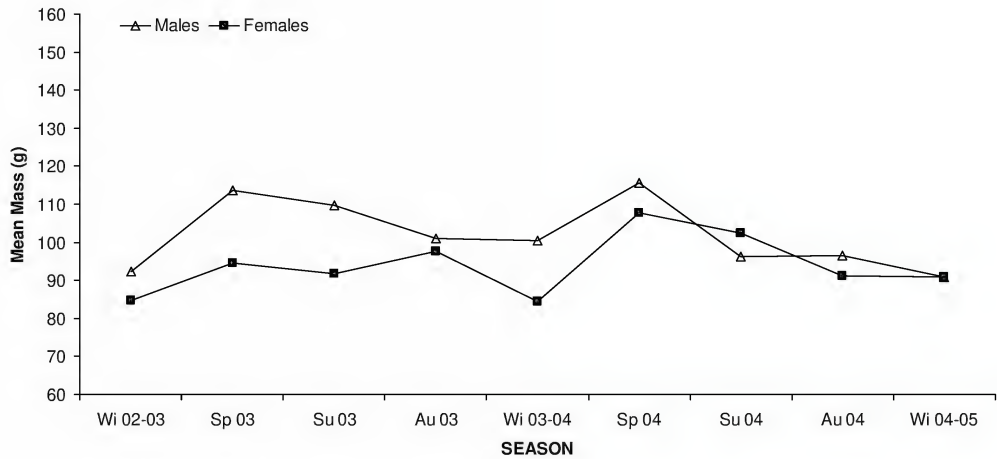
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FIGURE 2. Mean masses (g) for sub-adult and adult male and female *Sigmodon hispidus* in the 4 seasons in Chesapeake, southeastern Virginia. Large increases in mean mass for both sexes were observed from winter into spring, followed by a decline in mean mass later. Pregnant females were excluded from analysis.

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PROCEEDINGS OF THE 95TH ANNUAL MEETING

Virginia Academy of Science
Virginia Commonwealth University
May 17-19, 2017

Contents

Agriculture, Forestry, and Aquaculture

Posters

SEEING IS BELIEVING: THE IMPORTANCE OF HANDS ON EXPERIENCES IN THE DIVERSE WORLD OF MODERN AGRICULTURE. R. Spencer Irby & Tim C. Durham, Agriculture Program, Ferrum College, Ferrum, VA 24088. Education can be delivered through auditory, visual, and kinesthetic modalities. However, there is often a disconnect between theory and practice in agricultural education, elevating the latter in importance. A hands-on approach

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allows students to better grasp abstract concepts. This bridges and actualizes the theories broached in class. Accordingly, courses such as AGS 218: Regional Experiences in Agriculture at Ferrum College are an intuitive and beneficial complement to an educational curriculum, particularly for students in the midst of major coursework. In the aforementioned course, students observed and participated in a wide variety of agricultural exercises within the Commonwealth, while gaining firsthand experiences in a nontraditional, open air classroom setting. The course also provided a platform for students to converse directly with individuals in industry employment sectors of interest, with a particular focus on alumni mentoring. Perhaps most importantly, the course challenged and contextualized student perspectives on the global dilemma of resource scarcity and a growing population.

Astronomy, Mathematics and Physics with Materials Sciences

K-KINGS IN PRODUCTS OF DIGRAPHS. Morgan Norge & Peter LaBarr, Virginia Commonwealth University, Richmond, VA 23284-2014. A k-king in a digraph D is a vertex that can reach every other vertex in D by a directed path of length at most k. We consider k-kings in the strong product of digraphs. In particular, we determine the relationship between k-kings in the strong product of digraphs and k-kings in the factors of the product.

STOPPING POWER OF LEAD FOR ATMOSPHERIC MUONS. B. Murphy, C. Barazandeh & W. Majewski, Div. of Mathematics, Science & Engineering, NVCC. Our muon experiment is very distinct from most other experiments, as it has but a single detector instead of two in stereo. We with our single detector may identify stopped muons by their unique decay signature. In addition to measuring the muon lifetime, our detector also was permitted to measure the kinetic energy distribution of the muons (up to 140 MeV) as they come to be stopped in our detector, and the energy spectrum of the electrons created in the decay. In our presentation, we present them as graphs and interpret their shape. Our detector measures properties of not only stoppable low-energy muons with energy below 140 MeV but also of those muons passing through, with average energy as high as 4 GeV. An important property we discovered about them is their flux: the number of cosmic ray muons that hit the Earth's surface per unit area per unit time. Muons make a major part of the natural background ionizing radiation, with the average value of about 10,000 muons per square meter per minute at the Earth's surface. Because they are highly penetrating, reaching down through a kilometer or so of rock, we measured the attenuation of their flux by different layers of lead shielding, up to 25 cm thickness, of lead plates and bricks. Our results are expressed in terms of the lead stopping power – the energy a muon loses as it passes through 1 cm of lead. AIP Grant.

MUON LIFETIME AND ITS STANDARD MODEL OF PARTICLE IMPLICATIONS. C. Barazandeh & W. Majewski, Div. of Mathematics, Science & Engineering, NVCC. The NOVA Chapter of the Society of Physics Students for years was using its Muon Detector to register the arrival and stopping of a muon as a voltage pulse as an investigation into different aspects of cosmic radiation. This may detect a second pulse after a time delay of about two microseconds, which occurs from the electron created in the decay. From the distribution of the individual muons' lifetimes, we found the mean muon lifetime at rest, in agreement with the accepted data. From this measured lifetime, we calculated the strength of the Weak Nuclear Force – one of the four fundamental forces of nature. Using an SM relation, we found from our totally non-electric

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experiment the value of the elementary electric charge e, and so the strength of the Electromagnetic Force. This turned out to be a number close (error 7.5%) to the value 1.6E -19 C which we see in our introductory physics textbooks! Finally, from our simple muon lifetime experiment, we found the vacuum expectation value v (error of 4% from the accepted value) of the recently discovered fundamental Higgs field. This field pervades all space and determines the masses of all known particles of the visible Universe, including the Higgs particle mass itself. This is an amazing result, showing how powerful and mathematically consistent is the Standard Model of Particles. Supported: EF Grant.

PHYSICS OF PROPULSION & LEVITATION OF SELF-DRIVEN ELECTRODYNAMIC WHEELS. Jorge Del Carpio, Vincent Cordrey, Brady Murphy, Sepehr Samiei & Walerian Majewski, Department of Physics, Northern Virginia Community College, Annandale, VA. The NOVA Society of Physics Students chapter constructed an electrodynamic wheel using a motorized bicycle wheel with a radius of 12 inches and 36 one-inch cube Nd magnets attached to the rim of the wheel with one inch wooden spacing blocks between magnets. The radial magnetic field on the outer rim of the wheel is maximized by arranging the magnets in a series of Halbach arrays. The wheel is being upgraded to 76 magnets directly in contact with one another, doubling both angular rotation of the magnetic field and the density of pole reversals. In this talk, we describe a method of mounting magnets on the wheel which overcomes the repulsion forces which can be up to 70 lbs. The rotation of a wheel with alternating magnetic poles along the rim creates a time-varying magnetic field in the vicinity of the rim. When a conductive metal track is immersed in this area of strong reversing magnetic flux, the time-dependent flux induces eddy currents in the track, generating both lift (levitation) and drag (propulsion) forces on the wheel's magnets which can be measured with force gauges. Measurements of the low-density interim 38 magnet non-Halbach state of the upgraded wheel showed partial levitation can be achieved with non-Halbach systems. Electrodynamic wheels may have applications in magnetic levitation (Maglev) transportation, since multiple electrodynamic wheels could be used on a vehicle to produce lift, traction, and guidance forces via simple rotation of the wheels above conductive tracks. (Supported by: Society of Physics Students Sigma Pi Sigma research grants and The Virginia Academy of Science.)

PHYSICS OF PROPULSION AND LEVITATION OF A SELF-DRIVEN ELECTROMAGNETIC WHEEL. Vincent Cordrey, Sepehr Samiei, Brady Murphy, Jorge D. Carpio & Walerian Majewski, Dept. Physics., Northern Virginia Community College., Annandale VA 22003. The NOVA Chapter of the Society of Physics Students constructed what is called an electrodynamic wheel using a motorized bicycle wheel with a radius of 12 inches and 36 one-inch cube Nd magnets attached to the rim of the wheel, with the number of magnets later increased to 76. We describe how we mount magnets on a wheel, despite repulsive forces up to 70 lb. The radial magnetic field on the outer rim of the wheel is maximized by arrangement in a series of Halbach arrays which amplify the field along the rim. The rotation of a magnetic wheel near a conducting non-magnetic plate creates a time-varying magnetic field that can produce eddy currents by induction, providing both levitation and propulsion, measured with force gauges. Measurements were taken at a variety of wheel speeds, and the results were compared to the existing theoretical predictions. Partial levitation was achieved with the current electrodynamic wheel. Electrodynamic wheels may have applications in the magnetic levitation-based transportation "Maglev", since multiple electrodynamic wheels could be used on a vehicle to

produce by the same levitation, propulsion and guidance forces over a conductive track. Our configuration of a plate suspended above the rotating wheel can serve also as a model of noncontact conveyance of conductive plates in electrodynamic conveyor belts.

INVESTIGATING THE EFFECT OF MIDDLE INFRARED RADIATION (MIR) ON STANDARD CAPACITORS. Kyle S. Wenger[1], Dr. Brian C. Utter[2] & Dr. Giovanna Scarel[1], [1]Department of Physics and Astronomy, James Madison University, 22807 and [2]Bucknell University, Lewisburg, PA 17837. This research is a further investigation into the interaction between infrared radiation and matter with the main goal being new energy sources. Specifically, we look into the effect on standard capacitors used in electronics and comparing results with the findings of our thermoelectric device (TEC). The results support the assertion of previous research that the TEC behaves like a capacitor with a certain capacitance.

MATHEMATICS UNDERLYING ROTATIONAL KINEMATICS. Eric M Montag & Roland L Bowles, Dept. of Physics., Randolph-Macon College, Ashland VA 23005. The problem of parameterizing the group of rotations in Euclidean 3-space has been of interest since 1776 when Euler first proved his famous rotation theorem. Euler's notion was extended by the work of Rodrigues (circa 1841), Hamilton (circa 1843), and Cayley (circa 1845). Rodrigues derived a formula that permits direct calculation under certain conditions, while Hamilton generalized the underlying mathematics by introducing the new algebra of Quaternions that enabled the parameterization of the rotation group. Meanwhile Cayley developed Cayley Transformations, a mapping between real skew-symmetric matrices and orthogonal matrices. This presentation focuses on methodologies for calculating orthogonal coordinate transformation matrices $A(t)$ between two frames of reference F_a and F_b, where F_b is attached to a rotating body and rotates with angular velocity $\omega(t)$ relative to F_a, which we may regard as fixed in space. These methods for calculating $A(t)$ are discussed in light of a physics-imposed kinematical constraint. Parameterizations of $A(t)$ are presented and discussed in the context of published research findings, indicating that no three-parameter system can produce a continuous, global and non-singular transformation, but that a four-parameter representation suffices.

MULTI-CHANNEL TEMPERATURE ACQUISITION APPARATUS DESIGNED FOR USE IN POLY-DISCIPLINE UNDERGRADUATE RESEARCH LABORATORIES. Ethan C. Ramirez, Department of Physics, University of Mary Washington, Fredericksburg VA, 22401. Acquisition of temperature variations as a function of time or external influence during an experimental procedure is paramount in the collection of meaningful datum. Therefore this device serves as a cornerstone in any experimental environment whether that be a quantum optics table or carefully controlled ecological survey. Utilizing standard issued resistive transducers, each of the eight channels has a tested temperature range of 5 to 60 degrees Celsius with the real-time temperature displayed on a 20-4 character customizable LCD display. The system is centered around the popular ATMega328P and ADS1115 16-bit analog to digital converter (ADC) communicating via I2C also featuring an on-board USB drive for program and documentation storage. Further, the nodes are zeroed with respect to an on-board calibrated transducer for maximum precision and have been tested for accuracy against a trusted meter.

DESIGN AND SIMULATION OF A POLARIZED PURE PHOTON SOURCE FOR COMPTON SCATTERING FROM SOLID POLARIZED TARGETS. Donal Day, Dustin Keller, Darshana

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Perera & Jixie Zhang, Department of Physics, University of Virginia, Charlottesville, VA 22904. Wide angle Compton scattering from polarized protons holds great promise: access to the generalized parton distribution functions H and E with different weighting and moments than in other hard exclusive processes, emphasizing the u-quarks and the valence region. Previously, experiments were proposed using bremsstrahlung from polarized electrons striking a radiator. Unfortunately, the mixed electron-γ beam limits the polarized target performance due to radiation damage and restricted luminosity owing to the heat load. We have designed a pure photon beam line by placing a dipole magnet after the radiator which deflects the electrons away from the target and into a local beam dump. This approach has many benefits which include an order of magnitude increase in the photon luminosity and unrestricted use of transversely polarized targets while preserving robust target performance. We will discuss the physics motivation, the design, simulation, and the radiological aspects of the local dump. Supported by DOE Grant Number DE-FG02-96ER40950.

FABRICATION OF A LARGE-AREA HIGH-EFFICIENCY COSMIC RAY VETO DETECTOR AT UVA. Robert C Group, Department of Physics, University of Virginia, Charlottesville, Virginia 22904. Since the discovery of the muon, particle physicists have carried out a series of experiments aimed at measuring flavor violation in charged-lepton interactions. To date, no such violation has been experimentally observed. However, the Mu2e experiment at Fermilab will search for the charged-lepton-flavor-violating process of coherent muon-to-electron conversion in the presence of a nucleus with a sensitivity four orders of magnitude beyond the current strongest limits. The goal of single-event sensitivity requires that all backgrounds must sum to significantly less than one event. One potential background is due to cosmic-ray muons producing an electron with signal characteristics within the Mu2e apparatus. The cosmic-ray-veto system of the Mu2e experiment is tasked with vetoing such cosmic-ray-induced backgrounds with high efficiency while inducing low dead time and while operating in the high-intensity environment of the Mu2e experiment. The UVA HEP group has been leading the effort to design and prototype the CRV and plan to build this detector on site over the next few years. Highlights of this effort including prototype performance as well as an overview of the Mu2e experiment will be presented.

COMPARISON OF METHODS FOR TESTING THE SYNCHROTRON SELF-COMPTON PROCESS FOR ANALYZING THE HIGH-ENERGY RADIATION FROM BLAZARS. Steven D. Bloom, Department of Physics and Astronomy, Hampden-Sydney College, Hampden Sydney, VA 23943. Two methods are compared for observationally constraining the synchrotron self-Compton model as applied to blazars. One such method uses the observed radio properties of a source to determine physical parameters and predict the high-energy spectrum. Another uses the observed high-energy and low energy spectrum to constrain physical parameters of the source and the overall spectral shape and level. As applied to blazar Markarian 421, it is shown that both methods show similar results. Since the methods only partly overlap, it may be promising in the future to use both methods on well observed sources to develop the best fitting models.

THE GRAPH BRAIN PROJECT. Craig Larson, Department of Mathematics and Applied Mathematics, Virginia Commonwealth University, Richmond, VA 23284-2014. Craig Larson and Nico Van Cleemput (Ghent University) have developed the CONJECTURING program which can produce conjectures in any area of mathematics. Graph Theory is defined in part by its published

concepts (invariants and properties), examples and counterexamples, theorems, and open problems. After a variety of preliminary experiments, we now intend to start adding all published graph theory examples, invariants and properties, together with all known bounds for the independence number of a graph (a widely-studied NP-hard graph invariant), in order to discover new bounds---not implied by any published bounds.

A REALISTIC LOOK AT STARSHIPS. John Pavco, Dept Physics & Astronomy, NOVA Annandale VA 22003-3796. The discovery of planets outside our solar system together with the rapid advances in technology provides the scientific motivation for astronomers and physicists to consider sending a starship containing a variety of sensors on an actual journey to these planets. Concepts that have addressed starship designs that are possible in the near future are addressed in this study. These concepts include nuclear pulse rockets which employ nuclear bombs detonated beneath the vehicle. The solar sail concept employs a highly reflective spacecraft that utilizes photons from the sun to propel it. Ion thrusters that use electrostatic or electromagnetic forces to accelerate a stream of ions are also considered. A space-based cyclotron that employs the same physical principals as the first cyclotron that was put into operation by Ernest Lawrence in 1932 is described. Acceleration is accomplished by passing a charge through the potential difference of a capacitor a large number of times. A space ark concept for manned starship flight was also discussed. The space ark will be a gigantic cylinder where the crew would live on the inside walls of the cylinder.

PUTTING THE GENIE BACK IN THE BOTTLE: THE SCIENCE OF NUCLEAR NON-PROLIFERATION. Gerard P. Gilfoyle, Dept. of Physics, University of Richmond, Richmond, VA 23173. Nuclear non-proliferation, the efforts to limit the use and spread of nuclear weapons, began with the first development of the atomic bomb in 1945. The methods to detect, monitor, and verify the construction, movement, and testing of nuclear weapons has evolved as both the technology and the international landscape (and the threat) changed over the last seventy years. In this talk I will develop the technical features of these devices that drive many of the non-proliferation efforts around the world. Next, I will focus on two central issues. (1) I will discuss the methods used to detect and characterize a nuclear explosion anywhere in the world by the International Monitoring System (IMS) of the Comprehensive Test Ban Treaty organization. Attention will be focused on the series of tests performed by North Korea since 2006 and the performance of the IMS. (2) I will then examine the illicit trade in nuclear materials and the technical challenges faced when transnational groups (i.e. terrorists) pursue the construction of a nuclear bomb. Assessments of the threat will be presented and some of the ways that threat is mitigated will be explored. I will spend some time at the end talking about the relationship between science and our society and draw some conclusions.

Posters

MODELING POPULATION DYNAMICS OF INCOHERENT AND COHERENT EXCITATION. Hannah R. Killian & Brandon J. Rozek, Department of Physics, University of Mary Washington, Fredericksburg VA. In the three level atomic system, incoherent excitation leads to approximately one third of the population transferred from the ground state to the uppermost excited state. The objective of this study was to maximize the population transfer to the highest energy level while minimizing the amount of population in the intermediate state. Using

the properties of coherent light in a two-photon system, theoretical calculations were performed with the time dependent Schrödinger equation. By exploiting the orthonormality of the wave function and applying the near resonance, dipole, and rotating wave approximations, a model was developed to show population dynamics for a 3-level Rb^{87} system. Population transfer is driven by delayed laser pulses, and the model demonstrated that to maximize transfer the pulses should be in a counterintuitive order where the second laser field is applied prior to the first. Nearly complete population transfer was achieved from the ground state to the excited state using this method of coherent excitation.

Biology with Microbiology and Molecular Biology

USING DNA METHYLATION DATA FOR COMPUTATIONAL INFERENCE OF REGULATORY ACTIVITY. John T. Lawson[1] & Nathan Sheffield[1,2], [1]Department of Biomedical Engineering, University of Virginia, Charlottesville VA 22908 and [2]Department of Public Health Sciences, University of Virginia School of Medicine, P.O.Box 800717, Charlottesville VA 22908. Advances in sequencing technologies have enabled the generation of higher resolution and higher coverage DNA methylation data. More information about the cell may be gleaned from this data but new methods are needed to do so. We have developed an R software package called Methylation-based Inference of Regulatory Activity (MIRA) that can infer regulatory activity from single-nucleotide-resolution, genome-scale DNA methylation data. Using a set of regions that correspond to some biological annotation such as transcription factor ChIP peaks, we aggregate methylation from those regions across the genome in order to get a single, summary methylation profile for those regions. A score is also generated based on the shape of the methylation profile, with the assumption that a deeper dip in the methylation profile is associated with more protein binding and open chromatin which can be markers of activity. By comparing the methylation profiles and scores for different samples, it is possible to infer which samples have more activity in the regions of interest. In summary, the MIRA package provides the means to infer systems level regulatory information from DNA methylation data, using the R programming language.

3D MODEL LINKING SPATIAL PATTERN STIFFNESS TO DECREASE LUNG FUNCTION IN IPF. John O. Sangobowale[1] & Jason M. Kinser[2], [1]School of Systems Biology, George Mason University, Manassas, VA 20110 and [2]Computational and Data Sciences, George Mason University, Fairfax VA 22030. Idiopathic pulmonary fibrosis (IPF) is a chronic, progressive disease of unknown etiology, that is characterized by the histopathological pattern of usual interstitial pneumonia. Despite the expanded knowledge on the pathogenesis of IPF, the mechanical dysfunction associated with the extent of remodeling is still fairly misunderstood. This study attempts to construct a probabilistic representation of a three-dimensional elastic model of the lung to investigate the onset of fibrosis. Given a predefined probability, a random isolated lesion of the alveolar wall, represented by a spring in the lattice model was subjected to fibrosis. Collagen deposition was simulated in the basilar subpleural, and peripheral regions of the model to mimic fibrosis of an IPF lung. The local lesions of fibrosis, represented by the stiffening of springs along a strain dependent walk, accounts for the excess deposition of extra cellular matrix and tissue remodeling as the lungs parenchymal is distorted. Regional deposition of collagen and the local onset of fibrosis is achieved by increasing the elastic constants of the springs at the site of initiation and the springs along a strain dependent walk of length N. As these incidents increase, we hypothesize that the spatial orientation of the model will begin to morph, and the overall

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stiffness of the model increase until there is a sharp decline in its mechanical properties. The proposed model hopes to determine why pathologic progression of IPF does not always follow the mechanical behavior associated with the disease.

GENE VARIANTS OF THE AGER PROTEIN IN INFLAMMATORY METABOLIC DISEASE. Gladys A. Shaw[1], Rohini Mehta[2], Munkhzul Otgonsuren[2], Aybike Birerdinc, [1,2] & Ancha V. Baranova[1,2], [1]School of Systems Biology, College of Science, George Mason University, Manassas, VA 20110 and [2]Betty and Guy Betty Center for integrated Research, Inova Health System, Falls Church, VA 22042. Advanced glycation end products (AGEs) are cross-linked, non-degradable aggregates of proteins, lipids and nucleic acids produced in the course of ageing and many aging associated chronic diseases. Additionally, the receptor for advanced glycation end products (AGER) is an important player in the pro-inflammatory pathway. Clinically, inflammatory metabolic diseases, such as diabetes, non-alcoholic fatty liver disease (NAFLD), and obesity, may be aided by a) Varying levels of AGER isoforms; b) Polymorphisms in AGER influencing ligand-receptor interaction or AGER levels; c) Variations in concentration of AGER's ligand, advanced glycation end products (AGE). These variations will alter inflammatory milieu within the human body. TaqMan qPCR was employed in genotyping of 4 inflammation-related SNPs located within the RAGE gene (Gly82Ser, G1704T, T-374A, and T-429C) in 340 obese patients. ELISA assays were used in the analysis of AGER protein isoforms in the serum of these patients. Statistical tests were used to determine whether a particular genotype/haplotype of the AGER locus produces significantly different levels of AGER protein. Correlations between levels of circulating AGE and prevalence of NAFLD in those with specific AGER genotypes were detected. Our findings may explain why only a subset of NAFLD patients progresses to NASH.

MOLECULAR EXAMINATION OF DNA IN RODLET CELLS OF TELEOST FISH. Brandon S. Hamel & Dr. David T. Gauthier. Dept. of Biological Sciences, Old Dominion University, Norfolk, VA 23529. Rodlet cells are an unusual cell type found only in the tissues of teleost fishes. While their cellular morphology initially led researchers to classify them as a distinct parasitic species, observations of their behavior and function have demonstrated characteristics less typical of an invader and more of an endogenous or symbiotic cell. In order to resolve this discrepancy, molecular techniques were employed to compare the DNA of a rodlet cell to that of its host. The first utilized Laser Capture Microdissection to precisely excise rodlet cells from fish tissue in order to amplify and sequence the cell's DNA. The sequences from rodlet cell enriched and nonenriched samples were analyzed with BLAST against the SILVA small ribosomal subunit database. While the nonenriched sample returned hits from primarily other teleost fish, the rodlet enriched sample returned many hits from fungi, particularly from the genus *Malassezia.* This fungus is a known part of human commensal flora and subsequent PCR testing was not definitive in ruling out contamination, leaving the results inconclusive at this time. The second approach will utilize *in-situ* hybridization with digoxin labelled probes specific to a conserved region of the fish 18S gene. Visualization of this DNA-DNA binding should provide a more definitive resolution as to the identity of the enigmatic rodlet cell. (Supported by: The Virginia Academy of Science, Old Dominion University Honors College, and the Research for Undergraduates in Math and Science [RUMS] Program).

MAPPING QUANTITATIVE TRAIT LOCI FOR VOLUNTARY ETHANOL CONSUMPTION IN DIVERSITY OUTCROSS MICE. Kristin M. Mignogna[1], Aaron R. Wolen[3], Daniel M. Gatti[4],

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& Michael F. Miles[2,1], [1] Virginia Institute for Psychiatric, Behavioral, and Statistical Genetics, Virginia Commonwealth University (VCU) Richmond, VA 23298, [2] Department of Pharmacology and Toxicology, VCU, Richmond, VA 23298, [3] Center for Clinical and Translational Research, VCU, Richmond, VA 23298, [4] The Jackson Laboratories, Bar Harbor, ME 04609. Previous studies have estimated to heritability of Alcohol Use Disorder (AUD) to be roughly 50%, and have identified Quantitative Trait Loci (QTL) (genetic region significantly correlated with a trait) for ethanol-related behaviors. However, traditional mouse models do not provide enough genetic or phenotypic variation to identify QTL with enough precision to identify a specific gene. Therefore, the Diversity Outcross mouse stock was created to provide a unique level of both genetic and behavioral variation for high-resolution QTL mapping for complex traits, such as ethanol-related phenotypes. Ethanol-related measures have not yet been studied in these mice. This study employs an Intermittent Ethanol Access paradigm with three-bottle choice (15% ethanol, 30% ethanol, and water) to map QTL for voluntary ethanol consumption in a preliminary sample of Diversity Outcross mice (n=196). Although none of the QTL were significant, likely due to lack of power, provisional QTL were identified on chromosomes 15 and 14 with confidence intervals sized < 0.3cM. The QTL with the strongest association (on chr15) was located in an intronic region of the FAM134b gene, which has been previously linked to neuronal degeneration. NIAAA P50 AA022537, P20 AA017828, and U01 AAO16667.

THE RELEVANCE OF NATURALLY OCCURRING MUTATIONS IN CIRCADIAN COMPONENTS FOR CELLULAR SIGNALING. Philip E Stauffer, Dept. of Biol., Virginia Tech, Blacksburg VA. The mammalian circadian clock is a cell autonomous molecular clock that oscillates over a period of roughly 24 hours thus regulating physiology and behavior through differential gene expression in concordance with the time of day. Interestingly, model organisms with perturbations in their normal clock due genomic alterations or abnormal feeding cycles exhibit various health issues including proliferative disorders. Accordingly, animals deficient in the core circadian gene, Period 2 (Per2), are arrhythmic and show an increased rate of tumorigenesis when exposed to irradiation. Our lab has elucidated a molecular rationale for this observation by identifying a cross talk mechanism between Per2 and a critical node for cellular response to genotoxic stress that relies on Per2 function. We have shown that under normal physiological conditions, Per2 binds to and stabilizes the tumor suppressor p53 by preventing p53's ubiquitination by its dominant negative regulator, the oncogene Mdm2. Upon induction, p53 is post-translationally modified, forms a tetrameric complex that binds to DNA and regulates transcription of its target genes. In addition, our lab has shown that for p53 to exert its transcriptional activity, it must disassociate from its complex with Per2. More recently, mutations that alter the affinity of the two proteins have been identified and the effects this has on p53's stability and transcriptional activity as well as the transcription of core clock genes have been characterized in immortal cell lines.

THE EFFECT OF MICROBUBBLES ON MAMMALIAN CELLS. Caleb C. J. Wyckoff, Enbo Yang, Christopher Osgood, & Shu Xiao, Frank Reidy Center for Bioelectrics, Old Dominion Univ., Norfolk VA 23508. Microbubble induced shearing and jetting from bubble collapse has been closely related to mild traumatic brain injury and viability of cells. Chinese hamster ovarian cells (CHOs) were grown on coverslips and were individually placed upon a slide connected to a novel bubble generator governed by a piezoelectric transducer. We placed the coverslips upon the chamber in order to observe, in real time, the interaction of ultrasound induced microbubbles on

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cell membranes and viability. Cells affected by microbubbles at ~10µm were shown to have the greatest amount of propidium iodide (PI) uptake. Larger bubbles appeared to have no direct influence on the uptake of PI. Additionally, three characteristics of microbubbles were observed as they made an impact on cell viability: bubble equilibrium, bubble collapse, and bubble shrinkage. Bubble equilibrium describes microbubbles that did not collapse but remained present amongst cells. Bubble collapse describes microbubbles that "burst" thus affecting a certain area of cells. Bubble shrinkage describes microbubbles that softly dissipate amongst cells. Upon learning more about the distinct characteristics of microbubbles relating to viability, we hope to move forward and work directly with neural cells derived from induced pluripotent stem cells. The project was supported by a grant from Office of Naval Research (grant number: 12606014561).

EFFECTS OF NANODIAMONDS ON BONE MARROW-DERIVED MACROPHAGES PRE- AND POST-DIFFERENTIATION. Maisoun E. Bani Hani & Christopher Osgood, Dept. of Biol., Old Dominion Univ., Norfolk VA 23529. Nanodiamonds (ND) are carbon-based nanomaterial which have been suggested for a wide variety of biomedical applications. Their intrinsic fluorescence and ability to conjugate with biomolecules made these particles a promising candidate for drug delivery and *in vivo* imaging and tracking. Several studies on different cell lines and animal models exposed to ND reported very low toxicity of ND *in vitro* and *in vivo*. However, the effects of ND on bone marrow-derived macrophages (BMDM) has not been investigated. I aimed to study the effects of ND on bone marrow cells before and after they differentiate into macrophages. ND-treated and untreated cells differentiated into macrophages within seven to ten days and did not appear to alter cell's morphology. In addition, I assessed expression of macrophages surface markers, CD11b and F4/80, and did not find a significant difference between the ND-treated and untreated cells. After differentiation, BMDM were able to uptake ND in a time- and dose-dependent manner as assessed by flow cytometry experiments. Confocal microscope was used to determine the subcellular localization of ND inside the cells and the images show that ND enter the cell's cytoplasm but not into the nucleus. MTS assays showed that the cell's viability was not significantly affected by ND treatment. Pretreated or untreated BMDM with ND were able to phagocytose Cascade blue-labeled dextran particles regardless of the ND concentration used. These results suggest that ND are not toxic to BMDM; exposure of bone marrow cells to ND did not affect their ability to differentiate into macrophages and they did not affect BMDM viability or phagocytic activity.

T CELLS EXPRESSING CHIMERIC PD1 RECEPTORS THAT CONTAIN A DAP10 COSTIMULATORY DOMAIN ARE A POTENTIAL TREATMENT FOR MULTIPLE TYPES OF CANCER. Kelsey Deal & Amorette E. Barber, Department of Biological and Environmental Sciences, Longwood University, Farmville, VA, 23909. CD8 T cells have evolved as one of the immune system's best defense mechanisms against tumors. However, some tumors accumulate enough mutations to evade T-cell detection. These mutations include downregulating proteins that a T cell would normally use to identify tumors. Despite this, most tumors express proteins called Programmed Death Ligands. Since PD ligands are expressed on most tumor types, and not many other cells, they are ideal targets for potential therapies. To enhance tumor recognition by T cells, chimeric antigen receptors (CAR) consisting of the Dap10 costimulatory and CD3ζ activation domains fused to PD1 (chPD1) receptors were created and expressed in T cells. The anti-tumor efficacy of chPD1 T cells against murine liver and colon cancers was measured. Of the cell lines tested, all expressed PD1 ligands, making them potential targets for chPD1 T cells. In addition,

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chPD1 T cells induced tumor cell lysis showing a significant increase when compared to wild type T cells. ELISA and LEGENDplex analysis then determined high secretion levels of pro-inflammatory cytokines IFNγ, TNFα, IL-2, GM-CSF, IL-17, and IL-21 and low levels of anti-inflammatory cytokines IL-10 and IL-5 in both tumor types. Tumor targeting efficacy of the CARs was then tested in vivo by measuring tumor burden and survival. Results determined a significant decrease in tumor burden as well as an increase in survival. Therefore, adoptive transfer of chPD1 T cells could be a novel therapeutic strategy to treat liver and colon cancer.

EXPRESSION OF MATRIX METALLOPROTEINASES IN *NAEGLERIA FOWLERI*. Charlton Lam[1], Melissa Jamerson[2], Guy Cabral[1] & Francine Marciano-Cabral[1], [1]Department of Microbiology and Immunology, Virginia Commonwealth University, Richmond, VA, 23219 and [2]Department of Clinical Laboratory Sciences, Virginia Commonwealth University, Richmond, VA, 23219. *Naegleria fowleri* is a free-living amoeba found in freshwater lakes and ponds that causes Primary Amoebic Meningoencephalitis (PAM), a rapidly fatal disease of the central nervous system (CNS). PAM occurs when amoebae attach to the nasal epithelium and invade the CNS. This invasion involves binding to, and degradation of, extracellular matrix (ECM) components. Matrix metalloproteinases (MMPs) have been described in protozoa, such as *Plasmodium falciparum*, *Trypanosoma brucei*, and *Balamuthia mandrillaris*, and have been linked to their increased motility and invasive capability. In addition, MMPs are often upregulated in tumorigenic cells and have been attributed as responsible for the metastasis of certain cancers. In the present study, *in vitro* experiments indicated that MMPs are linked functionally to the ECM degradation process. Gelatin zymography demonstrated gelatinolytic activity in *N. fowleri* whole cell lysates, conditioned media, and media collected from *in vitro* invasion assays. Western immunoblotting confirmed the presence of the metalloproteinases MMP-2 (gelatinase A), MMP-9 (gelatinase B), and MMP-14 (membrane type-1 matrix metalloproteinase [MT1-MMP]). Hghly pathogenic mouse-passaged amoebae expressed higher levels of MMPs than the weakly pathogenic axenically grown amoebae. The functional relevance of MMPs in degradation of ECM components was confirmed through the use of MMP inhibitors, including 1,10-phenanthroline and ND-336.

SEX DIFFERENCES IN THE RESPONSE OF BDNF AND CRH IN THE BNST TO SOCIAL STRESS: IN THE NEUROBIOLOGICAL MECHANISMS OF ANXIETY. Hannah M Belski, University of Mary Washington. Anxiety is a complex, behavioral disorder that can devastate a person's life. While the causes of anxiety are complex and rooted in multiple brain and hormonal systems, a wealth of evidence points to a limbic brain region known as the bed nucleus of the stria terminalis (BNST) as a seat for many of the neurophysiological processes that contribute to this disorder. Furthermore, expression of the protein brain derived neurotrophic factor (BDNF) in the BNST is highly correlated with the expression of anxious behavior. While it is clear that the prevalence of anxiety disorder is higher in females, specific sex differences in the expression of BDNF in the BNST are unclear. Alongside BDNF, corticotropin releasing hormone (CRH) is an important modulator of behavioral and physiological responses to stress. I propose an experiment using a mouse model of social stress, a potent instigator of anxiety, to investigate the role that BDNF and CRH in the BNST might play in the sex differences that are observed in anxiety. I predict that both male and female mice will exhibit elevated anxiety following social stress and that this trend will be exaggerated in females. I also expect that BDNF and CRH expression in BNST will exhibit parallel patterns. While this experiment is still in progress, I have found so far

that subordinate animals show distinctly different behavioral patterns than the dominant mice in the colony. Our data also suggests more hierarchical stability among female cohorts compared to males.

Posters

CHARACTERIZATION OF A PUTATIVE KINASE ASSOCIATED WITH RESCUE OF *CRYPTOCOCCUS NEOFORMANS* PYK1Δ GLUCOSE UTILIZATION. Nicolas Terreri, Joshua Sellwood, & Michael S. Price, Dept. of Biology & Chemistry, Liberty University, Lynchburg, VA 24515. *Cryptococcus neoformans* persists in the central nervous system (CNS) via the utilization of carbon sources mainly from sugars like glucose. In a prior study assessing the role of glycolysis in CNS persistence by *C. neoformans*, we observed the delayed appearance of colonies on glucose-containing medium inoculated with the *pyk1Δ* strain. The colonies appeared as one of three morphotypes: filamentous, pseudohyphal, or yeast. Additionally, the rescue mutants displayed a temperature-sensitivity phenotype. Increased filamentation has been observed in *C. neoformans* with overexpression of the mating (MAT) locus. Therefore, we believe that the gene(s) responsible for rescue is located near the MAT locus of *C. neoformans*. Due to the morphology phenotypes observed for these *pyk1Δ* rescue mutants, we hypothesize that a partial genome duplication near chromosome 5 may be responsible for rescue of the *pyk1Δ* mutant on glucose-containing media. Two putative kinase genes located near the MAT locus were cloned into a vector containing the histone H3 promoter to drive constitutive expression; these constructs were introduced into the *pyk1Δ* strain and assessed for both glucose rescue and temperature sensitivity. This study describes the genetic capabilities available to pathogens to overcome severe metabolic disruption.

EFFECTS OF FIBRONECTIN FIBRIL ASSEMBLY ON CELL MOTILITY IN A BREAST CANCER PROGRESSION MODEL. Mary A. Anderson[1], Brian Griffin[1], Jennifer Koblinski, Ph.D[2], & Christopher A. Lemmon, Ph.D[1], [1]Virginia Commonwealth University, Virginia Commonwealth University Massey Cancer Center. Increased fibronectin fibril assembly and migration have been linked to epithelial to mesenchymal transition (EMT), a crucial step in cancer progression. The effects of the extracellular matrix on migration have not been thoroughly explored. In this study, we examine the changes in migration of 4 isogeneic cell lines of increasing malignancy in the presence of TGF-β, a soluble factor that induces EMT, and a fibronectin fibril assembly inhibitor, termed FUD. We found, as expected, TGF-β did stimulate migration; however, this effect is nullified by the addition of FUD. Addition of FUD without TGF-β had negligible effects on migration. The more malignant cell lines did not respond as drastically to the addition of either TGF-β or FUD. This was attributed to a lack of dependence on external signaling in the more malignant cell lines. This study shows possible applications of FUD as a method to return developing malignant cells to a more premalignant migratory state, as well as reduce the fibrotic build up that occurs in tumor formation, if used in the early stages of cancer progression. (This work was supported by the National Institute of General Medical Sciences of the National Institutes of Health under award number R01GM115678 (MPI: Lemmon and Weinberg)).

ROLE OF βNAC/ICD-1 IN CONTROL OF CANCER-ASSOCIATED GENE EXPRESSION IN *C. ELEGANS.* Marina Barmanova & Dr.Tim A. Bloss, Department of Biology, James Madison University, 800 S Main St, Harrisonburg, VA, 22801. Cells constantly undergo misfolded protein stress, thus triggering unfolded protein response (UPR), which can decrease misfolded protein

levels or initiate apoptosis depending on stress levels. The nascent polypeptide-associated complex (NAC) is heterodimeric chaperone that prevents misfolded protein stress in the ER; depletion of the NAC triggers a strong UPR. When not in complex, the individual subunits of the NAC are thought to regulate gene expression; in particular, the beta subunit of the NAC (βNAC) is thought to affect the expression of Epherin receptor 2 (EPHB2), a gene associated with cancer. I want to determine the putative regulation of the EPHB2 homologue Vab-1 by the ☐NAC homologue ICD-1 in *C. elegans*, with the hopes of gaining insight into the roles βNAC and EPHB2 in the development of cancer. The gonad of *C. elegans* provides a cancer cell model, and was characterized in *vab-1* knockouts with wild type and depleted levels of ICD-1. The depletion of ICD-1 from *vab-1* knockout worms generated mutant gonad phenotypes that differ from *vab-1* knockouts alone: primarily the growth of the gonad past the vulval midline of the worm, indicating continued gonadal growth throughout adulthood, abnormal for a wild type *C. elegans*. This phenotypic difference between treated and untreated vab-1 knockouts indicates ICD-1 may not be the only factor controlling the expression of Vab-1 in *C. elegans*.

CONSTRUCTION OF AN IPTG INDUCIBLE BETA-LACTAMASE EXPRESSION PLASMID FOR *STAPHYLOCOCCUS AUREUS*. Caitlyn R. Bishop & Gail E. Christie, Dept. of Microbiology and Immunology, Virginia Commonwealth University School of Medicine, Richmond VA 23298. The goal of this experiment was to construct a plasmid carrying the *blaZ* gene that is easy to assay in *Staphylococcus aureus*, and then examine, in both the suppressor strain and wild type *S. aureus*, how much the gene is expressed when a specific mutation of interest in the ribosomal binding site is present. First, a plasmid miniprep was done to isolate plasmid DNA from an overnight culture of *Escherichia coli.* To digest the plasmid DNA, a restriction enzyme was used to cleave the plasmid, resulting in the vector used for the subsequent insertion of the *blaZ* gene. For the polymerase chain reaction (PCR), we used two primers flanking the *blaZ* gene and a thermocycler to amplify the gene of interest. The DNA fragment from the PCR reaction was inserted into the vector using Gibson assembly (the Clontech In-Fusion® kit), resulting in the new plasmid. *E. coli* cells were transformed by electroporation, and the plasmid construct was verified by PCR and by direct DNA sequencing. The new plasmid was then introduced into a competent laboratory strain of *S. aureus* by electroporation. The resulting cells were infected with a generalized transducing bacteriophage and the lysate from this infection was used to transfer the plasmid to two recipient *S. aureus* strains, S2 and S66, by transduction. In conclusion, our study resulted in the successful construction of the plasmid containing the gene of interest and its introduction into the desired host strains for further analysis.

USING CRISPR/CAS9 GENOME EDITING TO STUDY NEURAL DEVELOPMENT. Lindsey N. Bixler & Gregory S. Walsh, Department of Biology, Virginia Commonwealth University, Richmond VA, 23284. CRISPR/Cas9 is a developing method in the genetic research field for genome editing. CRISPR (clustered regularly interspaced short palindromic repeats) is a method that bacteria and archaea use for adaptive immunity against foreign DNA. We take advantage of this method to test gene function by injecting guide RNA (gRNA) sequences that will cause disruption of normal gene expression through targeted DNA cleavage by the Cas9 DNA endonuclease. Computer programs are used to develop guide RNA sequences. Lab work includes PCR, gel electrophoresis, gel extraction, gRNA in vitro transcription, embryo injection, bacterial cloning, T7E1 assay, and plasmid sequencing. As proof of principle, we tested whether CRISPR/Cas9 editing of *frizzled3a* (*fzd3a*) affects axon pathway finding in zebrafish. We tested

two different *fzd3a* gRNA along with mRNA for Cas9. We found that *fzd3a* gRNA1 caused axon pathway finding errors. Roughly 15% of the axons traveled posteriorly, incorrectly. Injection of *fzd3a* gRNA2 had no visible defects underscoring the importance of testing multiple gRNAs. T7E1 assays confirmed that gRNA1 was more efficient than gRNA2 at inducing mutations at the fzd3a locus. Our data demonstrates that *fzd3a* is necessary for axon pathway of commissural neurons. CRISPR/Cas9 generates insertion or deletion (indel) mutations. We determined the nature of mutations induced by Cas9 using a gRNA targeted to the *tbx20* locus. PCR was used to amplify the *tbx20* locus from DNA extracted from F2 embryos with homozygous indel mutations in *tbx20*. PCR amplicons were cloned into plasmids, grown in bacteria, and sequenced. We found 3 novel deletion mutations in the F2 generation, demonstrating the usefulness of CRISPR/Cas9 for generating loss of function alleles. Future experiments will use CRISPR/Cas9 genome editing to test novel genes for their role in neural development.

O-GLCNAC TRANSFERASE MAY AFFECT THE ACTIVITY OF SUCCINATE DEHYDROGENASE. Joel A. Brenny, Emily R. Berguson, Grace Kim, and Dr. Pei Zhang, Department of Biology and Chemistry, Liberty University, Lynchburg VA. 24502. O-linked N-acetylglucosamine Transferase (OGT) is an enzyme that glycosylates proteins on Ser/Thr residues. We are exploring whether this modification occurs on Succinate Dehydrogenase (SDH) and what effect it may have on SDH activity. This will be investigated by mutating target sites on SDH, transfection of mutated SDH into a cell line, and performing an enzymatic assay. There is evidence that O-GlcNAc modification in cells plays a role in diabetes. Overexpression of OGT in muscle and fat cells has also been shown to cause hyperleptinemia and lead to diabetes pathology. This project will shed light on some of the fundamental mechanisms involved with metabolic disorders including these.

CHIMERIC PD1- EXPRESSING T CELLS AS A POTENTIAL TREATMENT OF OVARIAN AND BREAST CANCER. Shane Crean & Amorette E. Barber, Department of Biological and Environmental Sciences, Longwood University, Farmville, VA, 23909. CD8 T cells are one of the best defense mechanisms against cancer. However, to destroy tumor cells, a CD8 T cell must recognize tumor-associated antigens. Tumor cells rapidly develop many mutations that resulting in evasion of the immune response. In spite of this, most tumor cells have expression of Programmed Death Ligands (PD ligands). The tumor cells have higher expression of PD ligands than normal cells making them an ideal target for potential therapies. The therapy that was used for this study is a novel chimeric antigen receptor (CAR). CARs are modified antigen receptors that use genetic engineering to replace the recognition domain of the T cell receptor. In this experiment a CAR was used with the PD1 receptor as the tumor-targeting domain attached to CD3 zeta activation and Dap10 costimulatory domains, called the chimeric-PD1 receptor (chPD1). Previously, chPD1 T cells effectively treated murine lymphoma and melanoma. In this study, the anti-tumor efficacy of chPD1 T cells against breast and ovarian cancers was tested. RT-PCR and Flow Cytometry were used to test for the expression of PD1 ligands in murine breast and ovarian tumor cells. The tumor cytotoxicity and the cytokine production of the chPD1 T cells were measured in comparison to wild type T cells. All tumor types tested in the study with chPD1 T cells had significantly increased tumor cell lysis, as well as high expression of inflammatory cytokines. Therefore, this study is a potential therapy for breast and ovarian cancer.

BIOINFORMATICS ANALYSIS OF THE MOLECULAR PROPERTIES AND INTERACTION BETWEEN HUMAN ADENOVIRUS TYPE 36 PROTEIN E4 ORF1 AND HOST GLUCOSE PROCESSING PATHWAYS. George I. Dimitrov & Anna V. Baranova, School of Systems Biology, George Mason Univ., Fairfax VA 22030. There is increasing evidence that infection with human adenovirus-36 (Adv36) is associated with obesity. Ad36 infection leads to increased glucose uptake in adipose and muscle tissues, reduced hepatic lipid accumulation and increases insulin sensitivity. Research has shown that the viral gene, E4 orf-1, is directly involved in formation of obesity. E4-orf1 has a PDZ-binding domain at its C-terminus which upregulates phosphatidylinositol 3-kinase (PI3K) pathway leading to adipogenesis. We aimed to examine *in silco* the protein protein interactions (PPI) between E4-orf1 and human proteins involved in PI3K pathway. Our interest is in discovering methods to treat obesity in humans and developing therapeutics for diabetes.

CHARACTERIZATION OF VEGETATIVE PHENOTYPES OF EARLY RESPONSE TO DEHYDRATION 10 (ERD-10) MUTANTS IN *ARABIDOPSIS THALIANA*. Renee E. Hilelson & April N. Wynn, Dept. of Biol., Univ. of Mary Washington, Fredericksburg VA 22401. We examined two mutant alleles of the EARLY RESPONSE TO DEHYDRATION 10 (*ERD 10)* gene, SALK_087789 and SALK_097255, to observe the vegetative and floral phenotypic differences between the two mutant alleles and wild type plants. Phenotypic characterization included percent germination, bolt development and gross morphology. We observed and characterized a clear disruption in the phyllotaxy of both the *erd 10* alleles. The phyllotaxy of plants, like Arabidopsis *thaliana,* is where axillary branches grow from the apical stem in a spiral pattern. However, in both the *erd 10* mutants we observed an irregularity in this spiral pattern; with the axillary branches and silique doubled up alongside one another. The most extreme phenotype we observed was two siliques growing out of the same node on the stem. These vegetative phenotypes indicate that *ERD 10* may play a role in the growth and structure of the vegetative organs in *Arabidopsis thaliana.* Additionally, previous studies have indicated that ERD10 and SEUSS (SEU), a transcriptional adaptor protein, may both play a role in carpel and ovule development (Wynn et.al, 2011). Crossing of *seu-3* with both *erd 10* alleles will allow for phenotypic characterization of the F2 double mutant for additive or synergistic phenotypes.

CHARACTERIZATION OF MISFOLDED PROTEIN STRSS INDUCERS IN THE DIFFERENT CELL TYPES OF *C. ELEGANS*. Taylor L. Hutchinson, Rana Ihsan, & Dr. Timothy A. Bloss, Dept. of Biol., James Madison Univ., Harrisonburg VA 22807. Various stressors can cause proteins to misfold and aggregate, often leading to cell damage and even cell death. To manage this stress, the cell employs mechanisms that mitigate misfolded protein levels, including the presence of chaperones, proteins that help other proteins fold properly. The nascent polypeptide-associated complex (NAC) is an a/b heterodimeric chaperone that assists in the folding and localization of nascent polypeptides to the Endoplasmic Reticulum (ER) during translation. Depletion of the NAC in *C. elegans* results in numerous cell-specific phenotypes, including a strong induction of the ER-specific unfolded protein response (UPR), which can either save the cell or initiate cell death by apoptosis. To better understand the nature of the stress induced in the ER when the NAC is depleted, we are comparing phenotypes seen in NAC-depleted *C. elegans* to those observed in worms treated with ER stressors with known mechanisms of action, i.e. thapsigargin and tunicamycin. We are observing stress effects in fluorescently marked hypodermal cells, specifically the viability and number of hypodermal cells in embryos. We will

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perform similar analyses in neurons to determine cell-specific responses to NAC depletion. Misregulation of the NAC has been associated with numerous diseases; a better understanding of the NAC's role in protein stress management may help us understand why.

COMPUTATIONAL ANALYSIS OF DNA METHYLATION AND ITS ASSOCIATION WITH THE PRESENCE OF VARYING LEVELS OF NAFLD IN A COHORT OF DIAGNOSED NAFLD PATIENTS. K. Y. Jeong[1,2], A. Birerdinc[1,2] & A. Baranova[1,2], [1]School of Systems Biology, George Mason University, Manassas VA 20110. [2]Betty and Guy Beatty Center for Integrated Research, Inova Health System, Falls Church VA 22042. Several recent lines of research have shown that in addition to a genetically predisposed profile, certain epigenetic factors play an important role in both the development and progression of certain chronic diseases such as NAFLD. Particularly for diseases under the umbrella of Metabolic Syndrome, it has been demonstrated that epigenetic as well as genetic factors play a crucial role in both the presence and severity of these comorbidities. This project focused on incorporating the epigenetic regulations found with the etiological pathways of chronic diseases. Using data analysis software R, differential methylation data of selected relevant genes from a dataset in GEO was extracted for further analysis and visualization. Another software, PathwayStudio, was used to map the protein pathways for the proteins encoded by the selected genes in investigate their relationship with each other as well as the role they may play in NAFLD. Utilizing these software programs, the relationship between DNA methylation and NAFLD was investigated to display how varying methylation state influences gene expression and thus protein interactions that may prove to be significant in NAFLD progression.

THE DETERMINATION OF GENTIC DIVERSITY AMONG BREWING *SCCHARMOYCES* STRAINS. Arjun V. Jogimahanti & Dr. Fernando Tenjo, Dept. of Biology, Virginia Commonwealth University, Richmond, VA, 23220. *Saccharomyces* is a genus that contains several different species of yeast. Many of these species can be combined or genetically altered to produce hybridized species. In order to determine the genomic similarities and differences among brewing yeast species, PCR fingerprinting, specifically using single primer analysis, and gel electrophoresis are used to amplify a region in the genome and compare the potential genomic differences among the species. Twelve commercially grown species were obtained and analyzed in this experiment using three different primers: M13, $(GTG)_5$, and $(GACA)_4$. The banding patterns of each species was compared to the bands produced by the model organism, *S. cerevisiae*. It was hypothesized that if the banding patterns of the commercially grown species match the banding patterns of *S. cerevisiae*, then the commercially sold yeasts are closely related to the known versions. Comparison of the banding patterns across the three primers revealed that *S. kudriavzevii* and *S. bayanus* produced bands that contained that same number of base pairs that *S. cerevisiae* produced. Thus, it was likely that these species are closely related to the model organism.

FORENSIC ANALYSIS OF BIOLOGICAL SAMPLES COLLECTED FROM A CIVIL WAR MEDICAL CENTER. Kristin N. Jones, Emily L. Thomas & J. Thomas McClintock, Department of Biology and Chemistry, Liberty University, Lynchburg, VA, 24502. Forensic DNA analysis has gained much notoriety over the past decade as a tool in human identification. DNA analysis has recently been used to investigate samples of historical significance. During the last battle of the Civil War (Sailor's Creek Battlefield, Rice, VA), the Hillsman House served as a medical and

surgical center for wounded soldiers treating approximately 358 Union and 161 Confederate soldiers over a twenty-four hour period. The prominent "bloodstains" on the floor under the single surgical table and two post-surgical beds provides evidence of the vast number of soldiers treated. These presumed bloodstains, which also found their way through the cracks in the wood floors onto the supporting floor joists, were collected in the Fall of 2016 and subjected to various classic and state-of-the-art techniques to demonstrate the presence of blood. To examine the genomic profiles, DNA was isolated from these samples, quantitated, amplified using the GlobalFiler™ PCR Amplification Kit, and subjected to capillary electrophoresis. The generation of partial and/or complete DNA profiles confirmed the presence of human DNA, as well as the ability of DNA profiling to reveal a part of history from a battle fought over 150 years ago.

PREFERENTIAL ANTITUMOR CYTOKINE PROFILE IS INDUCED BY DAP10 CYTOKINE RECEPTOR IN MURINE CD8 T CELLS. Hailey Kintz & Amorette E. Barber, Department of Biological and Environmental Sciences, Longwood University, Farmville, VA, 23909. CD8 T cells play a key role in controlling cancer cells. However, when tumor cells evade recognition the immune system requires a boost to effectively combat cancer. Adoptive T cell therapy redirects T cells by genetic engineering to specifically target tumor cells. These novel therapies boost the immune system and leave healthy cells undamaged. Within the tumor microenvironment, a broad range of tumor cells exert immune-inhibitory mechanisms by the upregulation of programed death-1 (PD-1). Targeting PD1 may provide tumor specificity, therefore, our laboratory developed a murine chimeric antigen receptor (CAR; chPD1) to further enhance anti-tumor efficacy. Current CAR therapies use various costimulatory domains, however, it is not known which costimulatory domain induces the optimal combination for anti-tumor immunity. For this purpose, we developed chimeric PD1 receptors (chPD1) that include CD3ζ and CD28, Dap10, 41BB, GITR, ICOS, or OX40 costimulatory domains to directly compare anti-tumor activity. This study also aimed to compare the effector functions induced by the costimulatory domains in natural receptors and in CARs. Differences were observed in T cell survival with greatest survival of chPD1-41BB. Additionally, differences in cytokine secretion such as pro-inflammatory, anti-inflammatory, or Th17 associated cytokines greatly varied depending on which costimulatory domain was present. ChPD1-Dap10 induces T cell survival, as well as preferential combination of pro-inflammatory and Th17 associated cytokines. Therefore, chPD1-Dap10 has the potential to be used as a novel therapy.

GENE EXPRESSION OF AMYLOID PRECURSOR PROTEIN DURING ZEBRAFISH EMBRYONIC DEVELOPMENT: DOSE-RESPONSE EFFECTS OF COPPER EXPOSURE. W. David Knight, Benjamin J. Miller, & Katie M. Wiens, Dept. of Molecular Biology and Chemistry, Christopher Newport Univ., Newport News VA 23601. Copper accumulation plays a significant role in the progression of Alzheimer's disease (AD). Metal toxicities, including copper, increase the expression of amyloid precursor protein (APP). When APP is present in large concentrations, cleavage leads to the development of amyloid-beta (Aβ) plaques. These plaques cause neuronal cell death, leading to the symptoms of AD. Treatments that regulate APP/Aβ are the focus of many studies aimed at preventing and treating AD. However, there is currently no *in vivo* model system that allows for rapid testing of compounds improving APP/Aβ regulation. Zebrafish are an excellent model system for investigating treatments for AD. Copper toxicity has been examined in zebrafish embryos and adults, yet no work has investigated the ability of copper to alter APP/Aβ activity in zebrafish. Zebrafish embryos were removed from their chorion at 24 hours post

fertilization (hpf) and treated with 0, 5, and 10 uM copper sulfate at 24, 48, or 72 hpf. Embryos were then collected at 72 or 96 hpf for qRT-PCR analysis. Early treatment of copper sulfate at 24 hpf led to significant embryo death in contrast to later treatments at 48 and 72 hpf. *appa* mRNA expression decreased from 72 to 96 hpf for control embryos, but no change was seen across the timepoint collections after treatment with copper. Future studies will include acute versus long-term treatments to further screen this model for potential use as an efficient, *in vivo* drug-screening model based on environmental induction of AD. (Supported by faculty development grants from CNU).

CAENORHABDITIS ELEGANS let-381/FOXF ACTS UPSTREAM OF *hnd-1/dHand* IN THE SOMATIC GONAD. Kayla Lopez-Alvillar, Jill Bettinger, & Laura Mathies. Virginia Commonwealth University. In *C. elegans,* two identical cell divisions result in two somatic gonadal progenitor cells (SGPs) and two head mesodermal cells (hmcs). The SGPs remain multipotent, and form all somatic tissues of the reproductive system. One hmc undergoes programmed cell death while the other terminally differentiates, ceasing all production of new cells. We use this cell fate decision as a model to understand cell differentiation and multipotency. There is sufficient evidence that *hnd-1* plays a role in the SGP/hmc cell fate decision. *hnd-1* SGP defects are the first documented among genes that control gonadogenesis and *hnd-1* SGPs express markers typical of their sister hmcs. However, *hnd-1* does not act alone; *let-381* has been observed in SGPs at the same time as *hnd-1* and *hnd-1* and *let-381* mutants show similar phenotypes. The *hnd-1* gene contains three potential LET-381 (FoxF) binding sites in its regulatory sequence. To determine whether these sites were required for *hnd-1* expression in SGPs, we mutated or deleted all three sites in a functional *hnd-1::GFP* reporter and inserted the modified reporters into the *C. elegans* genome as single copy insertions. We report that the FoxF deletion results in two fewer GFP expressing cells when compared with the wild-type reporter, while the FoxF mutation results in variable loss of GFP expression in one or two cells. Future experiments will determine which cells require the FoxF sites for *hnd*-1 expression. NSF grant IOS-1314144 funded this work.

ALTERED MITOCHONDRIAL DNA MASS IN HEPATOCYTES IN RESPONSE TO FATTY ACID SUPPLEMENTATION. Peter M. Masschelin[1,2], Aybike Birerdinc[1,2], Rohini Mehta[2], Ancha Baranova[1,2], [1]School of Systems Biology, George Mason University, Fairfax, VA, 22030, [2]Beatty Liver and Obesity Research Program, Inova Health Systems, Falls Church, VA, 22042. Mitochondrial DNA copy number is a measure of both mitochondrial activity as well as overall health. As mitochondria can modulate their genome copy number in response to organelle stress and energy requirements, elucidation into changes in this number provides insight into diseases including Non-Alcoholic Fatty Liver Disease. HepG2 cells were exposed to 800uM and 250uM of three dietary fatty acids; Oleic, Palmitic, and Butyric Acid, for 2, 4, 6, 12 or 24 hours. mtDNA and nDNA specific primers enabled quantification of a ratio of mitochondrial to nuclear DNA, giving insight into the mitochondrial activity as well as mitochondrial health. Interestingly, exposure to a monounsaturated fatty acid, Oleic Acid, revealed no significant changes in the mtDNA/nDNA ratio for either exposure duration or concentration. Palmitic Acid, a saturated fatty acid, significantly altered the mtDNA/nDNA ratio only over time course (p=0.0361). Comparison of these results to exposure with a short-chain fatty acid, Butyric Acid, revealed no significant alterations following changes in either exposure duration or concentration. As saturated fatty acids have been linked to a variety of diseases, the observed changes in the ratio of mitochondrial to

nuclear DNA reveal a dynamic interplay of mitochondrial genome alterations to mitigate the detrimental effects of saturated fatty acids.

TRAFFICKING AND IDENTITY OF THE NGF-TRKA SIGNALING ENDOSOME AND ITS ROLE IN POST-SYNAPTIC DENSITY FORMATION. Kate McDaniel[1], Kelly Barford[2,3], Christopher Deppmann[4], Bettina Winckler[2,3], [1]UVa Undergraduate Neuroscience Major, [2] UVa Neuroscience Graduate Program, [3]UVa Department of Cell Biology, [4]UVa Department of Biology. Sympathetic neurons, responsible for the "fight or flight" response, have periods of growth and cell death during development that are regulated by the amount of available nerve growth factor (NGF). NGF is released by peripheral organs and functions as a signal that signals to growing neurons that they've reached their final destination. Upon the neurons reaching their final destination, NGF binds to its high affinity receptor, TrkA. TrkA and NGF internalize into an endosome which traffics retrogradely back to the soma to prevent apoptosis. While it is known that NGF-TrkA signaling endosomes (SEs) move retrogradely back to the soma, the Rab protein(s) associated with the SEs in the soma and beyond are unknown although multiple Rab proteins have been implicated. It has been shown that TrkA SEs are present in the dendrites of superior cervical ganglia (SCG) in mice and regulate post-synaptic density (PSD) formation. Additionally, if the MEK/MAPK signaling pathway is inhibited, TrkA SEs are not found in the dendrites and PSDs are not formed. Currently, we are investigating the pathway and identity of TrkA SEs to elucidate the route through which SEs arrive in the dendrites, the Rab protein(s) associated with the SEs at each point in that route, and the signaling pathways that are required for TrkA SEs to be present in the dendrites to regulate PSD formation. (Supported by: Double Hoo Award).

DEVELOPING NEW MOLECULAR BIOLOGY TOOLS TO STUDY THE *BACTEROIDES THETAIOTAOMICRON* OXIDATIVE STRESS RESPONSE. Arielle S. Montague, B. Ross Belvin, Nicai Zollar & Janina P. Lewis, Phillips Institute for Oral Health Research, Virginia Commonwealth University, Richmond VA 23298-0566. *Bacteroides thetaiotaomicron* is our bacteria of interest. It's a key member of the gut microbiome. In order to survive in the gut microbiome, it must have an oxidative stress response. We were able to grow *B. thetaiotaomicron,* an anaerobic organism, and we found that it is able to survive under high concentrations of hydrogen peroxide. In response, we created a plasmid system to express a florescent reporter gene whenever reactive oxygen species is present. We synthesized the *ahcp* promoter upstream of a fluorescence reporter gene. This construct was cloned into the Pg108 shuttle vector. Our goal is to develop new tools to help us understand various regulatory mechanisms, such as oxidative stress response.

T CELL COSTIMULATION ENHANCES ANTI-TUMOR IMMUNITY THROUGH CYTOKINE SECRETION AND TUMOR CELL KILLING. Emily A. Nylen & Amorette E. Barber, Department of Biological and Environmental Sciences, Longwood University, Farmville, VA, 23909. Chimeric antigen receptors (CAR) can be engineered for cancer therapy to enhance tumor recognition by T cells. One receptor that is a prospective target for a new CAR is PD1 because the ligands for the PD1 receptor are expressed on many cancer types. We created a chimeric PD1 receptor that consists of the extracellular domain of PD1 and the activating domain of CD3ζ. In addition, costimulatory domains CD28, Dap10, 41BB, GITR, ICOS, or OX40 were included. To determine which chPD1 receptor induced superior anti-tumor immunity, the efficacy of chPD1 T cells was measured. Previously it was determined that murine lymphoma and

melanoma cell lines express the ligands for PD1, thus making them ideal tumor targets to test the efficacy of the chPD1 T cells. Regardless of the costimulatory domain in the CAR, all of the chPD1 T cells induced similar levels of T cell proliferation and tumor cell lysis. However, differences were observed in the cytokine secretion profiles depending on which costimulatory receptor was included in the CAR. While most of the chPD1 T cell receptor combinations secreted both pro-inflammatory (IFNγ, TNFα, IL-2, GM-CSF, IL-17, and IL-21) and anti-inflammatory cytokines (IL-10) as determined by ELISA and LegendPlex analysis, chPD1 T cells containing a Dap10 costimulatory domain secreted high levels of pro-inflammatory cytokines, but did not secrete a significant amount of anti-inflammatory cytokines. Therefore, inclusion of the Dap10 costimulatory domain in the chPD1 receptor may induce the strongest anti-tumor immune response.

CHIMERIC PD1-EXPRESSING T CELLS AS A POTENTIAL TREATMENT FOR PANCREATIC AND KIDNEY CANCERS. Geoffrey Parriott & Amorette E. Barber, Department of Biological and Environmental Sciences, Longwood University, Farmville, VA, 23909. CD8 T cells are one of the immune system's best defense mechanisms against tumors. However, some tumors accumulate enough mutations to evade T-cell detection. These mutations include downregulating proteins that a T cell would normally use to identify tumors. Despite this, most tumors express proteins called Programmed Death 1 (PD1) Ligands. Since PD1 ligands are expressed on most tumor types, and not many other cells, they are ideal targets for potential therapies. One such therapy is the development of chimeric antigen receptors (CAR). CARs are modified receptors that use genetic engineering to replace the normal signaling domain with a different one. In this experiment, we used a CAR with the PD1 receptor as the tumor-targeting domain attached to CD3 zeta activation and Dap10 costimulatory domains, called chimeric-PD1 receptor (chPD1). Previously, chPD1 T cells were shown to effectively treat murine models of lymphoma and melanoma. The purpose of the study was to test the anti-tumor efficacy of chPD1 T cells against murine kidney and pancreatic cancers. Flow cytometry was used to determine that all kidney and pancreatic tumor cell lines tested expressed the PD1 ligands. Therefore, these tumor cells were potential targets for our chPD1 T cells. ChPD1 T cells also had increased lysis of the tumor cells compared to wild type T cells. In addition, chPD1 T cells also secreted more pro-inflammatory cytokines, including IFN-γ, TNF-α, and GM-CSF, but did not secrete the anti-inflammatory cytokine IL-10. Therefore, chPD1 T cells could be a novel therapeutic strategy to treat pancreatic and kidney cancer.

MURINE PROSTATE AND BLADDER CANCER CELLS ARE KILLED BY CHIMERIC PD1 EXPRESSING T CELLS. Elle Richardson & Amorette E. Barber, Department of Biological and Environmental Sciences, Longwood University, Farmville, VA, 23909. Cancer is the second highest cause of death in the United States. Treatments such as chemotherapy and radiation have shown some promising results for patients, yet can be are detrimental to healthy tissues. Over the past decade scientists have been targeting the immune system to fight off cancer. Many cancers are able to mutate proteins on the surface of their cells to be undetected by T cells. By isolating a patient's T cells scientists can genetically alter these cells into chimeric antigen receptors (CAR) that specifically target tumor cells. CARs are genetically engineered receptors that replace a normal signaling domain with a new one that is more specific. These CARs are then administered back to the patient, where they specifically bind and kill cancer cells. Specifically, our lab uses a CAR with the PD1 (chPD1) receptor as the tumor targeting domain. In this experiment, we tested

the efficacy of chPD1 T cells against murine Prostate and Bladder cancer. Flow cytometry was used to determine whether PD1 ligands were expressed on all cancer cells. We then cocultured wild type (wtPD1) and chimeric (chPD1) T cells with tumor cells and measured cytotoxicity. For all four cancer types tested, chPD1 T cells significantly killed tumor cells. Hence, chPD1 could be an ideal treatment for multiple types of cancer.

OPTIMIZATION OF HPLC TECHNIQUE FOR THE QUANTIFICATION OF 2'-DEOXYCYTIDINE ANALOGS IN DNA SAMPLES. Caroline E. Roberts, Dr. Gary D. Isaacs, & Dr. Greg Raner, Department of Biology and Chemistry, Liberty University, Lynchburg, VA, 24515. Epigenetic regulators are capable of modifying gene expression in response to environmental inputs. Cytosine (C) methylation is one such regulator, which has been linked to various health issues, including birth defects, Alzheimer's disease, and cancer. Following methylation, C residues may undergo demethylation by the action of the ten-eleven translocation (TET) family of enzymes. TET enzymes oxidize methyl cytosine (5mC), leading to the sequential formation of hydroxymethyl cytosine (5hmC), formyl cytosine (5fC), and carboxy cytosine (5caC). Discovery of the demethylation pathway has led to the identification of 5hmC as an epigenetic modifier within neuronal gene bodies. Studies have also found that 5fC and 5caC are important regulators in the regeneration of unmethylated C. Because of this, one must differentiate between the five variants of C when determining methylation status. Previously, high-performance liquid chromatography (HPLC) was used for the quantification of C methylation but only to identify C and 5mC concentrations. The purpose of this study was to establish an HPLC protocol for the quantification of all C analogs involved in the methylation pathway with the ultimate goal of analyzing epigenetic modifications related to dietary folate inputs. When a standard mix of all five variants of C was analyzed using published protocol for the separation of C and 5mC, there was significant overlap between C and 5hmC peaks. By increasing the pH of the running buffer and changing from a C18 to a phenyl-hexyl column, a protocol was established that allows for separation and quantification of all analogs of C. Partial funding for this project was provided through the Liberty University Alumni Research Scholarship.

GENE EXPRESSION CHANGES ASSOCIATED WITH ALZHEIMER'S DISEASE. Christopher G. Schreiner, Dept. of Biol. & Chem., Liberty Univ., Lynchburg VA. 24515. Genetic analysis techniques were used to determine gene expression changes associated with Alzheimer's disease. DNA and RNA samples were isolated from control CD1 mice and transgenic Alzheimer's mice. The DNA and RNA were run on a gene sequencer, and the data collected were run through the DNA Subway to generate a list of the top 20 genes that had the largest expression changes between the CD1 and Alzheimer's models. Eight of these genes were chosen for secondary confirmation via qPCR based on their underrepresentation in the literature and possible Alzheimer's links: Gm26924, Lyz2, Gm4204, Gpnmb, Actr3, Gfap, Gm5506, and Gm26880. Of these 8, 3 were confirmed via qPCR to have increased expression levels in the Alzheimer's mice: Lyz2, Gpnmb, and Gfap. The qPCR results for the other 5 genes showed either little to no expression change, or a change opposite that seen in the data from DNA sequencing. More research will need to be performed to determine the function of the 3 confirmed genes and to determine why there was conflicting data between the qPCR and DNA sequencing analysis for the other 5.

EFFECTS OF FOLATE ON LEARNING AND MEMORY IN MICE: TIMING AND TYPE OF FOLATE HAS DIFFERING LONG TERM RESULTS. Caleb Schreiner, Christopher Schreiner,

Caroline Roberts, & Gary D. Isaacs, Department of Biology & Chemistry, Liberty University, Lynchburg VA 24515. Folic acid (FA) is a common artificial form of dietary folate which is especially important for pregnant mothers to prevent neural tube defects in newborns. The biologically active form of folate that is also commonly found in nature is 5-Methyltetrahydrafolate (5-MTHF). As many Americans had low folate diets the government mandated that certain foods be reinforced with folic acid such as bread and cereals. This study compares the memory and learning capabilities of mice in four different folate diets. The mice were either started on folic acid and then given a folate deficient diet shortly after they were weaned, or had a constant diet of folic acid, or were started on 5-MTHF and then given a folate deficient diet shortly after they were weaned, or had a constant diet of 5-MTHF. Y-maze, Novel Object, and Barnes Maze tests were performed on the mice. The Y-Maze showed no differences between the diets. The novel object maze showed a non-significant trend that the mice on the 5-MTHF deficient diet outperformed the corresponding mice on the FA deficient diet. The Barnes maze demonstrated with some significance that the mice on the 5-MTHF and 5-MTHF def diets outperformed their FA counterparts. An additional novel object test at 18 months may strengthen the current results.

EXAMINING THE EFFECTS OF SULFUR-REDUCING BACTERIA ON BIOREMEDIATION OF CONTRARY CREEK. Periklis Smith & Lynn O. Lewis, University of Mary Washington. This project proposes a method to lower the impact of acid mine drainage (AMD) in ecologically affected areas surrounding abandoned mine sites by adding a carbon source to reverse the impact on affected streams and rivers. My research exploited the only organisms that can live in these toxic conditions by changing what they use as an energy source. The mining site I observed mined pyrite. Sulfate is a major byproduct of pyrite and contributor to AMD. Addition of low cost treatments consisting of 10g/L of leaf clutter or 5 g/L of unsulfured molasses were used to test changes to the effectiveness of the bioremediation process. Carbon source treatments were carried out for 43 days, and were subjected to temperatures of 5.5 or 25 degrees Celsius. Each of the two treatment types at corresponding temperatures saw increases in pH levels after the 43 day treatment. pH levels were 3.486, 3.421, and 3.340 for control, leaf clutter, and molasses treatments respectively at 5.5 degrees Celsius at day one. A pH of 3.331, 3.323, and 3.134 for control, leaf clutter, and molasses treatments respectively was found at 25 degrees Celsius on day one. After 43 days all treatments saw a significant increase in pH. The pH level increased to 4.570 and 4.632 for leaf clutter and molasses treatments respectively at exposures to 5.5 degrees Celsius. There was a pH level increase to 5.995 and 6.594 with leaf clutter and molasses treatments respectively at exposures to 25 degrees Celsius. The increase in pH levels from exposure to alternate carbon sources supports the effects of carbon on microbial activity as a possible bioremediation process for AMD.

CARBAPENEM AND OTHER BETA-LACTAM RESISTANCE IN FRANCISELLA. Cody Waisanen & Monique L. van Hoek, School of Systems Biology, George Mason Univ., Manassas VA 20110. *Francisella* (*F.*) *tularensis i*s the causative agent of the disease tularemia (Rabbit fever) and is considered a biothreat agent. In light of emerging bacterial resistance to many antibiotics, and carbapenems in particular, we sought to examine the role of carbapenem and beta-lactam resistance in *Francisella.* Minimum inhibitory concentration (MIC) testing was conducted on a BSL-2 mode strain of *Francisella* as well as mutants which contained transposon insertion mutations within genes that were either potentially β-lactamases as well as one which was

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annotated as a zinc-binding protein as a control. The MICs for *F. novicida*, against a variety of β-lactam based antibiotics were compared to the MICs for the insertion mutants. Tests indicated that one of the mutants had increased sensitivity to several of the carbapenems tested, and increased sensitivity to the mono-bactam tested. This experiment used *F. novicida*, and has only tested carbapenems and a mono-bactam so far. The wide range of activity for this gene, and the lack of sensitivity in mutants of the other putative β-lactamase gene demonstrates that our results are in agreement with results already published, that the *Francisella* genome encodes for only one functional β-lactamase protein.

Biomedical and General Engineering

FLOW IN DETERMINING PULMONARY ENDOTHELIAL PROGENITOR DIFFERENTIATION. Patrick A. Link & Laszlo Farkas & Rebecca L. Heise, VCU. In vascular fibrotic diseases such as pulmonary hypertension, endothelial cells (ECs) containing subpopulations of endothelial progenitor cells (EPCs) are exposed to dramatic changes in fluid flow and shear stress. We hypothesized that altering flow rate will shift the phenotype of clonally expanded EPCs toward EnMT, decreasing angiogenic potential. We show here that EPCs retain their phenotypic plasticity under conditions of low flow or low turbulence, but lose their angiogenic potential when exposed to high shear, turbulent flow. EPCs were isolated from the lungs of Sprague-Dawley rats by sequential positive (EC and progenitor marker) and negative selection (hematopoieitic marker), followed by repeated clonal enrichment of highly proliferative cells. Cells were cultured in a C2025 hollow fiber bioreactor (FiberCell) for 48 hours under varying flow rates at which point the cells were detached and subjected to a matrigel angiogenesis assay for 24 hours. After 24 hours, the cells were stained with Calcein AM and imaged for angiogenesis. Images were taken and quantified using AngioTool (National Cancer Institute). EPCs from the turbulent region of the bioreactor lost the ability to undergo angiogenesis more than the laminar regions. Cells from the high flow rates experiments lost the ability to undergo angiogenesis more than their low flow counterparts. The EPCs from low-flow, laminar region maintained significantly more angiogenic potential than the high-flow, turbulent regions (p<0.05). These results suggest EPCs may contribute to the propagation of vascular diseases such as pulmonary hypertension by responding to varying vascular flow.

ANTIBACTERIAL BEHAVIOR OF PROTEIN NANOPARTICLES DERIVED FROM PIG LUNG EXTRACELLULAR MATRIX. Gabrielle M. Cotman, Patrick A. Link, Robert A. Pouliot, & Rebecca L. Heise, Virginia Commonwealth University Richmond, VA Protein nanoparticles are an effective carrier for pulmonary drug delivery, due to safe degradation products and the ability to avoid impaction and sedimentation. Pulmonary drug carriers also must overcome biofilm produced by bacteria, so nanoparticles must be effectively anti-microbial. The objective of this study was to engineer protein nanoparticles derived from porcine lung extracellular matrix that are stable and reduce bacterial growth. Pig lung was decellularized, lyophilized, and cryomilled into powder which was digested in acetic acid and nanoparticles were retrieved through electrospray deposition of the digest. *E. Coli* was plated on a 96 well plate in LB media with pig lung ECM nanoparticles at increasing concentrations for 6h on a shaker at 37°C. The optical density of the samples were measured at 600 nm wavelength. Results showed that 0.5 mg/ml protein nanoparticles reduced average bacteria biomass by approximately 27%. To conclude, as nanoparticle concentration increased, the amount of bacterial growth decreased.

IMPROVING THE DESIGN OF PEPTIDE MEDIATED NANO DRUG-DELIVERY FOR TUMOR TARGETING IN BRAIN CANCER PATIENTS. Neeha Gambhirrao VCU Honors Virginia Commonwealth Univ., Richmond 23384. Nanotechnology can provide scientists with a valuable set of research tools and help develop advanced clinical devices. By considering current research in nanoparticle construction and the biology of brain tumors, it can be concluded that effective construction of nanoparticle drug delivery for brain cancer involves targeting specific pathways and proteins such as the ubiquitin and P13k.To make these nanoparticles even more effective, their surfaces are decorated with molecules that guide them to the tumor site by allowing the nanoparticle to interact with the tumor cells and release the drug it is carrying. Because the P13K and ubiquitin pathways are mainly mutated during the presence of brain tumors, the effective construction of nanoparticle drug delivery for brain cancer involves the targeting of these specific pathways. a new approach for targeting these proteins to the CNS across the blood-brain barrier is required for future treatments of neural conditions such as cancer. Nanoparticle carriers can address several drug delivery problems which could not be effectively solved in the past and include reduction of multi-drug resistance effects and penetration of the blood-brain-barrier.

A COMPUTATIONAL STUDY OF THE KINEMATICS AND JOINT MECHANICS OF FEMOROACETABULAR IMPINGEMENT. Brandon K. Marine & Jennifer S. Wayne Ph.D., Dept. of BME, Virginia Commonwealth Univ., Richmond VA. 23284. Patients presenting with femoroacetabular impingement (FAI) have abnormal contact between the femoral neck and acetabular rim leading to surrounding tissue damage as well as progressive levels of pain in daily use. Predicting this impingement as well as simulating its resolution through surgical correction would provide important information preoperatively. This study utilizes computational modeling to investigate hip kinematics during a sit-to-stand transfer and changes occurring with FAI. A combination of rigid bony anatomy and force vectors representing soft tissue structures were used. Ligaments were given a tension-only behavior based on their stiffness and muscle force magnitudes were designated based on the phase or time of motion. Kinematics predicted by the simulations agree well with experimental evidence during the initial phases of this activity; refinement of control parameters continues. Once further validated, a disease-state model of FAI will be utilized for comparison. Quantification of the FAI model relative to the healthy anatomy was done through a known measurement called the alpha angle, the angle between the femoral neck axis and that of the femoral head/neck transition. The FAI modeled has an alpha angle of 65.2 degrees while the healthy model has an alpha angle of 53.7 degrees. Joint angles of the trunk, pelvis, and hip are recorded with respect to time during the simulations and will be compared to quantify this alteration in motion.

AUTOMATIC CALCULATION OF FEMORAL VERSION USING PROXIMAL FEMUR LANDMARKS. Nathan J Veilleux & Jennifer S Wayne, Department of Biomed. Eng., Virginia Commonwealth Univ., Richmond VA. 23220. The success of total hip replacement surgery depends on proper anteversion of the femoral stem. Femoral version is defined as the angle between the femoral neck axis and distal condyles (transcondylar axis) in a horizontal plane. An excessively retroverted or anteverted femoral stem will lead to impingement and stem loosening, shortening the life of the hip implant. However, there is no accurate way for surgeons to measure native femoral version, since the femoral condyles aren't visible during the surgery. This means that a new technique for estimating version that only uses the proximal femur (the region visible

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during surgery) needs to be developed. CT scan data for 80 entire femurs and 215 proximal femurs has been provided by the VCU Department of Radiology. Each CT scan has been converted into a 3D model in the image processing software Mimics (Materialise, Leuven Belgium). The point cloud from each model was subsequently extracted and imported into Matlab (The Mathworks, Natick MA), where a program has been developed to automatically calculate femoral version using the entire femur. The program has been adapted to estimate femoral version using measures within the proximal femur. Six axes on the proximal femur that are visible intraoperatively have been defined. The angle between each candidate axis and the femoral neck axis will be used to assess their utility as surrogate measures of femoral version by comparing consistency to that calculated from the entire femur. The ultimate goal of this project is to create a tool that can be used to assist orthopaedic surgeons in aligning the stem of a femur implant during total joint hip arthroplasty.

AUTOMATED ANALYSIS OF ACETABULAR SURFACE FEATURES FROM CT SCANS. Patrick A. Jones, Nathan J. Veilleux & Jennifer S. Wayne, Department of Biomedical Engineering, Virginia Commonwealth University, Richmond, VA 23220. The complex morphology of the pelvis has made it challenging to automate computational methods for identifying features of this structure. More accurate and descriptive modeling of the pelvis, and in particular the acetabulum, would improve patient outcomes by providing additional preoperative planning tools for diagnosing conditions of the hip and planning interventions. This study builds upon previous work in which Higgins et al. developed a computational tool for isolating the rim of the acetabulum from Computerized Tomography (CT) images; we have developed additional methods for automatically determining acetabular surface features from CT scans. Using MATLAB (MathWorks, Natick MA) to correctly orient and search point clouds created from hemipelvis CT scans, we were able to automate the process of identifying the points of the interior surface of the acetabulum. From these points of the acetabulum, those that describe an interiorly placed sphere were identified and subsequently used to generate a best fit sphere. The accuracy of the fit was assessed through a root mean square error, validating this method for automatically determining asphericity of the acetabulum.

ANALYSIS OF STRESS TRANSFER IN THE HIP DURING SIT TO STAND. Morgan T. Kimmel, Brandon K. Marine & Jennifer S. Wayne, Ph.D., Dept. of Biomedical Engineering, Virginia Commonwealth University, Richmond VA 23220. Osteoarthritis (OA) of the hip occurs for multiple reasons, all resulting in the destruction of articular cartilage. One influential factor is excessive stress experienced in the joint over time during various activities of daily living. The sit-to-stand activity frequently generates pain in such patients. Simulating this activity in a finite-element-analysis (FEA) can provide an understanding of stress generation in the hip cartilage and the potential for OA progression. A CT scan of the hip provided by VCU Radiology was segmented in MIMICS (Materialise, Leuven Belgium) and then imported into Solidworks (Dassault Systems, Waltham MA). Then 3 models were constructed: (1) sitting: 90° flexion of hip, (2) intermediate: 135° flexion, and (3) standing: 180° flexion. 90%, 190%, and 100% of a 750 N bodyweight were applied to the hip joint as the force experienced in the specific positions. Cortical bone and articular cartilage were modeled as linearly elastic and isotropic with moduli of 16 GPa and 0.4MPa, and Poisson's ratios of 0.29 and 0.1, respectively. The stress distribution will be compared between the 3 positions, focusing on the location, area of contact, and peak magnitudes of contact stress. This approach can be used to simulate hip pathologies and results compared to the healthy joint.

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SENSITIVITY OF EYE TRACKING TASKS TO MILD TRAUMATIC BRAIN INJURY. Mary A. Kannan & Paul A. Wetzel, Dept. of Biomedical Engineering, Virginia Commonwealth University, Richmond VA, 23220. Persistent and chronic post concussive symptoms following a mild traumatic brain injury have been well-documented. Standard exams may not show all the deviations from normal and patients are often unaware of changes in cognitive ability. Eye movements can open a window into individual brain function and impairment. For this study, 71 post-concussive participants and 75 normative population participants were enrolled by The Brain Injury and Mechanisms of Action of Hyperbaric Oxygen for Persistent Post-Concussive Symptoms after Mild Traumatic Brain Injury Study (BIMA). The Eyelink II eye tracking system recorded eye movements at 500 Hz. Participants were subjected to various eye movement tasks, designed to produce responses from the saccadic and smooth pursuit systems, which are controlled by different parts of the brain and utilize different control pathways. Significant differences were found between the mTBI and control groups in the smooth pursuit tasks. The horizontal ramp task showed that the mTBI group made less saccades during the task, but the saccades were larger in amplitude and overall fixation time was greater ($p < 0.001$), indicating that they had difficulty making smooth pursuit movements. These differences indicate that smooth pursuit tasks could be sensitive to tracking recovery in mTBI.

Posters

CELLULAR ENDOPLASMIC RETICULUM STRESS AND CYTOKINE RESPONSE IN AGE-ASSOCIATED EXPERIMENTAL VENTILATOR INDUCED LUNG INJURY. Franck J. Kamga Gninzeko, Michael S. Valentine, Joseph A. Herbert, Matthew B. Schneck, Rebecca L. Heise, Department of Biomedical Engineering, Virginia Commonwealth University, Richmond, VA 23236. Mechanical ventilation is a necessary clinical intervention; however, it can result in ventilator induced lung injury (VILI). The largest population requiring mechanical ventilation is the elderly, and age is a predictive factor in the severity of VILI. However, the exact relationship between age and VILI is unknown. There is also evidence that Endoplasmic reticulum (ER) stress increases with age (1). Both in vivo and in vitro data obtained via staining (CHOP and TUNEL) and gene expressions of primary Type 2 alveolar cells (AT2) from old static and stretched cells at 15% the surface area for 24 hours were analyzed. The results suggest that age associated increases in ER stress upregulate the severity of VILI, and that ER stress inhibition (via the used of 4-PBA) can attenuate these effects. Furthermore, ER stress significantly contributes to the inflammatory response mediated by cytokine signaling (Ccl2, Ccl6, Ccl7, Ccl9, Csf1, Cxcl10rb, Il1a, and Il2rg), thus activating and recruiting macrophages at the injury sites produced by mechanical ventilation. (Supported by: The National Institute of Health)

MECHANICAL FORCE ACROSS E-CADHERIN REGULATES EPITHELIAL ACINI HOMEOSTASIS. Vani Narayanan & Daniel E. Conway, Dept. of Biomedical Engineering, Virginia Commonwealth University, Richmond, VA. Epithelial cells, when placed into 3D matrix, spontaneously form spheres with a single central lumen, known as acini. A number of studies have shown that these three dimensional structures behave differently when compared to 2D monolayers of cells. Using an existing E-cadherin FRET-force biosensor, we observed that E-cadherin force was significantly increased in acini as compared to monolayers. CFTR-induced lumen pressure was found to be a major generator of increased cell-cell force in 3D acini. Using

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low- and no-force mutants of E-cadherin, we observed that changes in E-cadherin force impaired the formation of a single-lumen, but was rescued by increasing lumen pressure. Additionally, increasing lumen pressure induced cell proliferation in an E-cadherin-dependent manner, likely similar to stretch-induced proliferation in 2D monolayers. Finally, we observed that EMT results in a decrease in E-cadherin force, and that maintaining increased lumen pressure was sufficient to block the TGF-β induced decrease in E-cadherin force as well as markers of EMT. Our results show that mechanical force across the adherens junction is an important regulator of 3D epithelial organization. Furthermore, we have identified osmotic-driven lumen pressure as a major contributor to mechanical forces in these structures. (Supported by: American Cancer Society Institutional Research Grant, NIH R35GM119617).

MECHANICAL FORCES APPLIED THROUGH THE NUCLEAR LINC COMPLEX. S.T. Newman, K. Bathula & D.E. Conway, Thomas Nelson CC & BME, VCU. Mechanical forces are transmitted across the cytoskeleton as both tensile and compressive forces. These forces are applied to organelles, such as the nucleus. The nucleus is directly connected to the cytoskeleton by the nuclear LINC (Linker of Nucleoskeleton and Cytoskeleton) complex. Recently our group used a novel force biosensor to show that the LINC complex is subject to actomyosin-generated tension. In our work, we developed methods towards answering two fundamental questions: 1) what proteins comprise the nuclear LINC complex and 2) what is the relationship between externally applied forces to the outside of the cell and LINC complex force. To identify additional components of the LINC complex we used a biotin proximity labeling technique, known as BioID, in which biotin ligase was inserted in to nesprin-2G, a LINC complex protein. Cells expressing this fusion protein were then exposed to biotin, and then all biotinylated proteins were purified using streptavidin beads. Several unique proteins were identified by mass spectrometry. Towards the later question, how the nucleus is coupled to the extracellular environment, we wanted to identify methods to allow for direct imaging of actin and the nucleus in living cells. We tested two approaches, Lifeact GFP, a peptide which associates with actin, and Chromobodies for actin and lamin, small antibodies that are conjugated to GFP. Chromobodies were found to provide higher resolution imaging of the actin cytoskeleton compared to Lifeact. (Supported by: Bridges to Baccalaureate R25GM102795, and the Thomas F. and Kate Miller Jeffress Memorial Trust)

THE EFFECT OF PIG LUNG EXTRACELLULAR MATRIX NANOPARTICLES ON CELLULAR PHENOTYPES. Alexandria M. Ritchie, Patrick A. Link, Michael S. Valentine & Rebecca L. Heise, Department of Biomedical Engineering, Va. Commonwealth Univ., Richmond VA 23284. Nanoparticles may have the ability to allow cells to repair damage to tissues. Nanoparticles derived from the extracellular matrix of pig lungs (PLECM) were used to study the impact on bone marrow-derived monocytes (BMDM) phenotypes of young, male,C57BL6 mice. Monocytes were isolated from the femur and tibia of the mice. M1 macrophages promote inflammation of tissue while M2 macrophages promote tissue regeneration. Cells were kept naïve, activated to become M1-like or M2-like, or had nanoparticles added to them (0.5mg/mL). The cells were activated by altering the growth media to include LPS and IFNy for M1 activation or IL-4 and IL-13 for M2. Activated cells were stained and run through the flow cytometer to determine whether or not the activated cells were exhibiting macrophage characteristics. Cells that exhibited markers +CDllc/-CD206 were designated as M1. Those that were -CD11c/+CD206 were designated as M2. Preliminary data suggests that BMDMs activated with PLECM nanoparticles have a high percentage of -CD11c/+CD206. Thus, nanoparticles from porcine lung

ECM may have the potential to induce BMDMs to become M2 macrophages and promote regeneration. (Supported by: The Civil, Mechanical and Manufacturing Innovation (CMMI) of the National Science Foundation CMMI under award number CAREER CMMI-13516).

RECELLULARIZATION OF DECELLULARIZED LUNG SCAFFOLDS IMPROVED BY EXTRACELLULAR MATRIX AIRWAY COATING AND PREFUSION CULTURE. Bethany M. Young, Robert A. Pouliot, & Rebecca L.Heise, Department of Biomedical Engineering, Virginia Commonwealth University, VA 23284. Decellularized tissue has shown great promise in the field of regenerative medicine for whole tissue engineering. The current technique uses chemicals to remove all cellular material that could cause a negative host response. These harsh chemicals can cause a major challenge because of disproportional stripping of vital extracellular matrix (ECM) proteins from the airway surfaces causing minimal cell coverage when recellularizing airways. We hypothesize that refunctionalizing the airway surface with ECM proteins in the form of a hydrogel will increase cell attachment and direct proper airway cell signaling. In this research, decellularized pig lung extracellular matrix (PLECM) hydrogels, previously designed by our lab, have been characterized and evaluated as a coating for the airways of a decellularized mouse lung. Hydrogel pepsin digestion time was optimized for mechanical properties, crosslinking, and cellular response. We assessed the type and amount of each key ECM protein present within the hydrogel. After hydrogel optimization, we designed, built, and tested a bioreactor for culture. We then quantified cell coverage through histology and cell proliferation, which showed the ECM hydrogels may be promising for recellularization. Improving recellularization techniques for complex organs will bring regenerative medicine closer to a realistic organ replacement strategy.

Botany

FOREST COMMUNITY RESPONSE TO THE REWETTING OF THE GREAT DISMAL SWAMP NATIONAL WILDLIFE REFUGE, VIRGINIA Stephen W. Bendele & Robert B. Atkinson, Dept. of Organismal and Environmental Biology, Christopher Newport University. Many coastal forested peatlands such as the Great Dismal Swamp were ditched and drained beginning in the colonial period. As a result, plant community composition has shifted to favor less hydrophytic species such as *Acer rubrum* and *Liquidambar styraciflua*. Increases in water levels occurred following the formation of the federally-administrated Great Dismal Swamp National Wildlife Refuge in 1974, but two recent fires (2008 and 2011) suggested that further increases in water levels were needed. Two water control structures (weirs) were installed in September of 2013 but the effect on water tables and plant communities was not known. The purpose of this study was to evaluate peatland water table and plant community and responses to the installation of weirs. Groundwater and vegetation were monitored at 15 forested stands for a three-year period. Mean water table depth increased approximately 25 cm following the installation of the weirs despite normal climatic conditions. Plant composition exhibited minimal shifts in importance values and prevalence indices, a form of weighted average. Future management should seek to increase water levels further, targeting a Seasonally Flooded-Saturated hydrologic regime. Funding was provided by the VA Field Office of the US Fish and Wildlife Service and by NC Parks.

FLORISTICS OF THE ABRAMS CREEK WETLANDS, WINCHESTER CITY AND FREDERICK COUNTY, VIRGINIA. Woodward S. Bousquet[1] & Gary P. Fleming[2], [1]Environmental Studies Program and Biology Department, Shenandoah University, Winchester, VA 22601, and [2]Natural Heritage Program, Virginia Department of Conservation and Recreation, 600 E. Main St., Richmond, VA 23219. Long known to naturalists as a bird-watching site, the botanical significance of the 24-hectare Abrams Creek Wetlands was first reported in 1980. A 1993 study by the Virginia Natural Heritage Program led to collaboration with Shenandoah University beginning in 1998. The present study (2010-2017) produced a complete floristic inventory, characterized natural communities, and examined factors shaping species composition and temporal dynamics. The 304 vascular plant species documented include 20 state-rare taxa. Natives constitute more than 70% of the species richness and over 80% of the vegetative cover, except in transition zones. Two state-rare communities are present: calcareous wet prairie and calcareous muck fen. While Winchester City has protected more than one-third of the site as the Abrams Creek Wetlands Preserve, the city's vegetation management plan for the preserve has not been implemented. A buffer zone has been proffered around a proposed adjacent development in Frederick County, but a new road will cross a portion of the wetlands as part of that project.

ANALYSIS OF THE EFFECT OF HYDROLOGY ON SOIL CARBON DIOXIDE EMISSIONS IN PEATLANDS IN SOUTHEASTERN VIRGINIA. Christina G. Mirda & Robert B. Atkinson, Dept. of Organismal and Environmental Biology, Christopher Newport University, Newport News, VA. 23606. Peatlands are characterized by deep accumulations of plant matter, which forms as a result of soil saturation and a lack of soil oxygen. Anoxia reduces soil microbial decomposition rates while still supporting primary production. The US Fish and Wildlife Service and the VA Department of Game and Inland Fisheries are installing several water control structures in forested habitats within the Cavalier Wildlife Management Area (CWMA) in southeastern Virginia in order to rehabilitate selected swamp attributes. The purpose of this experiment is to determine the effect of three hydrologic treatments (fully saturated, mostly-saturated, and moist-no water added) on soil carbon emissions. We obtained 24 soil cores from 4 sites, 6 cores per site, at CWMA. CO_2 emission rates of the soil cores were measured every day for 22 days with a LICOR. The fully-saturated and mostly-saturated treatments exhibited higher carbon emissions than the moist treatment. These findings suggest that a partial restoration of peatland hydrology at CWMA may inadvertently increase soil carbon emissions.

INVESTIGATIONS INTO THE NATURAL HISTORY AND REPRODUCTIVE BIOLOGY OF *BOECHERA SEROTINA* (BRASSICACEAE). Conley K. McMullen[1] & Paul J. Harmon[2], [1]Department of Biology, James Madison University, Harrisonburg, VA 22807 and [2]Wildlife Diversity Program, Natural Heritage Group, Wildlife Resources Section, West Virginia Division of Natural Resources, Elkins, WV 26241. *Boechera serotina* (Brassicaceae), known by the common name of shale barren rock cress, is a federally endangered plant restricted to Devonian shale barrens and adjacent shale slope woodlands in West Virginia and Virginia. It is considered endangered due to its limited natural habitat, as well as loss of habitat from road development, grazing by livestock and canopy closure through succession. The site of this study, the Little Fork Shale Barren in Pendleton County, WV, is on property owned by the U. S. Navy. This plant is a facultative biennial, perhaps better described as a short-lived, monocarpic perennial. Ongoing studies by the authors are documenting the development stages of this species via a combination of time-lapse photography and direct observations. Pollination studies, flower visitor

observations, and nectar sampling are part of the studies being conducted. The potential effect of herbivory on this species is also being examined. Here we report on research conducted in 2014-2015. Results of these experiments are currently being analyzed. This study was funded in part by Cooperative Agreement No. N62470-13-2-8026 between the U. S. Department of the Navy and the West Virginia Division of Natural Resources. Additional funding was provided by James Madison University.

THE FLORA OF VIRGINIA PROJECT: A 2016-2017 PROGRESS UPDATE. Marion B. Lobstein, Professor Emeritus, Northern Virginia Community College. The Foundation of the Flora of Virginia Project (FFVP) continues to make solid progress on development of the mobile version of *Flora of Virginia* Application or App. In March and in April of 2017, App prototypes for Android and iOS mobile devices were released to volunteer testers. Tester feedback on the prototypes is now being submitted. The first full version of the App is scheduled for release the weekend of September 29, 2017 at the tristate meeting of the Virginia, Maryland, and West Virginia Native Plant Societies. The development of a graphic key for use by the general public as well as inclusion of the dichotomous keys from the *Flora*, descriptions at family, genus, and species levels as well as background reference material from the *Flora* is in final stages of development for inclusion in the release of the first version of the App. Continued support of the *Flora of Virginia* project efforts by VAS and the VAS Fellows, resulted in contributions of a total of $4000 in 2016. A version of a *Flora* App for both Macintosh and personal computers will the next Foundation of the Flora of Virginia Project.

DENDROECOLOGICAL ANALYSIS OF ATLANTIC WHITE CEDAR AS A PREDICTOR OF HISTORIC HYDROLOGIC CONDITIONS IN MID-ATLANTIC PEATLANDS. Robert B. Atkinson & Abigail H. Weaver. Dept. of Organismal and Environmental Biol., Christopher Newport Univ. Atlantic White Cedar (AWC) (*Chamaecyparis thyoides)* was once a dominant species in the Great Dismal Swamp (GDS), but AWC swamps are now an endangered ecosystem due to altered hydrologic regimes. Recent fires have been unnaturally destructive and land managers have begun to install weirs to raise water levels in the GDS, but minimal historic water level information exists to guide those efforts. The purpose of this study was to evaluate tree ring growth patterns in AWC in two reference sites for which a 14-month hydrograph classified the hydrologic regime as temporarily flooded (TF) and seasonally flooded, saturated (SFS). Tree cores from 54 trees per site were obtained and correlations were attempted with Palmer Drought Severity Index (PDSI) values obtained from the National Oceanic and Atmospheric Administration. Trees were sanded with progressively finer sand paper, cross-dated, measured to 0.001 mm, and were converted to basal area. Mean tree annual growth in the TF site in GDS ($1,100$ mm^2) was greater than in the SFS AR site (240 mm^2) and growth response to PDSI at the two sites diverged. Trees in GDS responded positively to PDSI (i.e., higher precipitation associated with higher growth, particularly in July of the current year) and AR trees exhibited a weakly non-significant negative response. Differences in AWC growth at two time scales (lifelong and annual) may help classify hydrology and aid restoration.

BEGINNINGS OF THE UNIVERSITY OF RICHMOND HERBARIUM: THE BOTANISTS AND THEIR COLLECTIONS. W. John Hayden, Dept. of Biol., Univ. of Richmond. The first herbarium at UR was assembled by P.R. Merriman in connection with his "*Flora of Richmond and Vicinity,*" published posthumously in 1930. Unfortunately, Merriman's collections were destroyed

by fire in 1925. Extant collections at URV began in earnest by R.F. Smart in the 1930's. Smart collected ca 2,400 specimens of fungi and myxomycetes; Smart also hosted Harvard botanist, M.L. Fernald, on numerous trips to southeast Virginia, receiving in return many duplicate specimens from the Gray Herbarium. Further, two of Smart's students, both from the class of 1937, are particularly noteworthy. E.S. Luttrell's ca 1200 specimens of Cladoniaceae are, perhaps his greatest contribution to URV, but he was a prolific collector of all groups of plants and fungi. J.C. Strickland, who returned to UR as a biology faculty member 1946-1979, collected ca 2,000 specimens, mostly cyanobacteria and other photosynthetic cryptogams. Ongoing digitization of URV has revealed additional specimens that pre-date URV's foundation by Smart in the 1930s. Two biology faculty members, J.T. Johnson and F.H. Wilson (plus Wilson's students), contributed slightly more than two hundred specimens, collected between 1925 and 1928. URV holds a total of six specimens, all acquired relatively recently, from the 19th C. The oldest specimen in the collection is *Corchorus siliquosus*, *Rugel 109*, collected in 1846 from Key West, Florida; it was received, via exchange, from the Institute of Jamaica (IJ).

ANATOMY AND MORPHOLOGY OF STAMINATE FLOWERS OF *ACALYPHA SETOSA* (EUPHORBIACEAE). Sarah Kwon & W. John Hayden, Dept. of Biol., Univ. of Richmond, Richmond, VA 23173. *Acalypha setosa* is a weedy annual plant native to the Neotropics. Like other members of the genus, *A. setosa*, forms three distinct flowers: staminate, pistillate, and allomorphic. This study focused on staminate flowers, studied by light (LM) and scanning electron microscopy (SEM) using standard techniques; because of their fragility and small size, staminate flowers were enclosed in microporous capsules during critical point drying for SEM. Specimens were greenhouse-grown. Staminate flowers are borne on short axillary spikes; flowering sequence within the spike is scattered. Staminate flowers open in the morning and shed their pollen well before noon. At anthesis staminate flowers are ca 0.75 mm wide and 1.00 long, including the pedicel; each pedicel is jointed at the position of post-anthesis abscission. The flower consists of four valvate sepals and eight stamens arising from a central column. Abaxially, sepal midveins bear prismatic crystals on short multicellular protuberances. At anthesis filaments reveal slight sclerification of cells on their upper (adaxial) surfaces. The bisporangiate anthers, often characterized simply as vermiform, are contorted irregularly while enclosed in the bud and dehisce in a unique helically twisted fashion at anthesis. The tapetum is amoeboid; at maturity, anther walls bear only remnants of epidermis and consist almost entirely of endothecial cells; middle layers are absent.

INDUSTRIAL HEMP PRODUCTION IN VIRGININA: CHALLENGES AND OPPORTUNITIES. Michael H. Renfroe, Dept. of Biol., James Madison Univ., Harrisonburg VA 22807. Virginia passed legislation allowing industrial hemp research through university-based research programs beginning in 2016. Because industrial hemp belongs to the species *Cannabis sativa* L., which is classified as a Schedule 1 controlled substance by federal legislation, there are considerable challenges to conducting this research. Registration with the Drug Enforcement Agency, along with obtaining a state Industrial Hemp Growers License was required to begin a research program. Seeds had to be imported, which involved a number of federal and foreign agencies. Despite these challenges, there are opportunities for the production of industrial hemp in Virginia. During 2016, an oil-seed cultivar was grown in two locations in Virginia. From this one cultivar, three products were developed. Seeds were harvested and pressed for oil. Oil was converted to biodiesel and used to operate farm equipment. Press cake was used for animal feed.

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Hemp stems were cut, dried, and baled. Stems were shredded in a bale shredder and used for animal bedding. Industrial hemp contains less than 0.3 percent tetrahydrocannabinol and has a different morphology and chemistry than marijuana. Further development of this crop, which has high potential in Virginia will be severely restricted until a better legal distinction is made between industrial hemp and marijuana and restrictions on crop production and product marketing are eased.

DENDROECOLOGY OF DISJUNCT *THUJA OCCIDENTALIS* L. FORESTS IN THE SHENANDOAH VALLEY, VIRGINIA. Joshua A. Kincaid, Environmental Studies Program, Shenandoah University. *T. occidentalis* (northern white cedar) is a long-lived and shade tolerant coniferous tree species of eastern North America. The range of northern white cedar extends across southeastern Canada to the Great Lakes and New England regions of the United States. Scattered disjunct populations exist as far south as North Carolina and Tennessee. This research represented a dendroecological analysis of two disjunct *T. occidentalis* forests located approximately 440 km south of the contiguous range margin of the species. While *T. occidentalis* was a dominant species at both sites, it was absent from the smallest size class. The age structure of *T. occidentalis* also suggested a lack of recruitment after 1940. The disturbance chronology was consistent with canopy gap-scale processes that generally affect a small number of trees each decade. Mean sensitivity of the *T. occidentalis* chronology was within the range of values reported for the species in northern portions of its range. The positive association between previous summer temperatures and *T. occidentalis* growth is unique to this study, but has been observed in other southern conifers. The relationship between growth and precipitation was in general agreement with studies from northern portions of the range. There was a consistent positive correlation between radial growth and PDSI.

NOTES FROM NATURE - PLANTS OF VIRGINIA: AN UPDATE ON THE ONGOING DIGITIZATION OF VIRGINIA'S HERBARIA AND CITIZEN-SCIENTIST PARTICIPATION. Andrea Weeks, Dept. of Biol., George Mason Univ., Fairfax, VA 22030. In 2014, 11 Virginian herbaria (GMUF, FARM, VPI, LYN, VMIL, URV, LFCC, JMUH, VCU, BDWR, AVCH) began creating publicly-accessible online databases of their specimens, including high-resolution digital images (www.sernecportal.org). Participation by undergraduate student imaging assistants and citizen-scientists has been integral to the project's success. To date, the project has expanded to include other herbaria and gathered 194,609 records of herbarium specimens. Images of the specimens are now routed through the online crowd-sourcing platform, NotesfromNature.org, that allows the public to transcribe specimen label information into database fields. Specimens are bundled into themed groups called, "Plants of Virginia" and have resulted in ca. 10,000 specimens being fully transcribed. Engagement of undergraduate students and members of the Virginia Master Naturalists and the Virginia Native Plant Society in the process has fostered substantive opportunities for communicating the importance of herbaria for research. Moreover, new modules for educating students about Virginia's flora and the emerging field of biodiversity informatics have been developed using the sernecportal.org database. (Supported by Virginia Native Plant Society and National Science Foundation, EF-1410086 "The Key to the Cabinets: Building and sustaining a research database for a global biodiversity hotspot.").

PLANT ANATOMY IN THE SERVICE OF PHARMACOGNOSY. Charles J. Gordner & W. John Hayden, Dept. of Biol., Univ. of Richmond, Richmond, VA 23173. Botany and medicine

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have deep and complexly intertwined roots. Since ancient times, plants have been recognized as conveniently available and effective sources of molecules to influence human physiology. Because of their reliance on plant-based medicines, for much of history across diverse cultures, physicians have needed to be skilled in pharmacognosy, the accurate identification of medicinal plants. The challenge of pharmacognosy starts with whole plants as encountered in the field, and extends to verification of the identity of previously collected (and often dried or powdered) single organs (roots, leaves, etc.). Since crude preparations of medicinal plants are dissociated from diagnostic reproductive structures, microscopic anatomical features are often required for determination of correct identity. Thus, plant anatomy and pharmacognosy also have intertwined roots. We studied secretory trichomes of three medicinal plants found in central Virginia via light and scanning electron microscopy. Although each of these plants produce medicinally important compounds in epidermal trichomes, details of their anatomical structure readily distinguish each. Secretory trichomes of *Dysphania ambrosioides*, are short-stalked and asymmetrical; those of *Pluchea camphorata* are similarly short-stalked, but symmetrical, and co-occur with elongate uniseriate non-glandular trichomes; and glandular trichomes of *Artemisia annua* are nearly sessile.

BIOTIZATION OF *DIONAEA MUSCIPULA* WITH *BURKHOLDERIA PHYTOFIRMANS*. Peter L. Riley & Michael H. Renfroe, Dept. of Biol., James Madison Univ., Harrisonburg VA 22807. Venus fly trap (*Dionaea muscipula*) is a carnivorous plant with a very restricted natural range and is threatened in the wild from illegal harvesting. Venus fly traps are commercially propagated by tissue culture, but have difficulty acclimatizing from in vitro conditions to in vivo growth in potting soil. Recent investigations have given more attention to the role of microbes in the rhizosphere, and especially non-pathogenic endophytic bacteria that promote plant growth and increase environmental hardiness and stress tolerance. The effect of *Burkholderia phytofirmans* strain PsJN was investigated for beneficial effects on acclimatization of Venus fly trap. Inoculations of Venus fly traps with bacteria in the tissue culture propagation phase proved detrimental to propagation on traditional sucrose based media. However, substitution of maltose ameliorated this effect. When Venus fly traps were acclimatized on sterile potting medium, the presence of endophytic bacteria made no significant difference in plant survival. However, when acclimatizing on non-sterile potting medium, presence of endophytic bacteria reduced mortality by 36% compared to non-inoculated control plants. This study demonstrates the need for further investigations of plant biotization during or following axenic propagation of plants through tissue culture and the importance of plant growth promoting bacteria in the rhizosphere and within plant tissues.

Posters

DEVELOPING HYDROLOGIC RESTERATION TARGETS FOR THE GREAT DISMAL SWAMP USING ATLANTIC WHITE CEDAR TREE RINGS. Alexandra Balzer, Abigail Weaver & Dr. Robert B. Atkinson Organismal and Environmental Biology Christopher Newport University, Newport News, VA A study of Atlantic White Cedar (AWC) Trees was conducted to create a tool that uses growth measurements (BAI) to characterize the historical hydrologic levels in the Great Dismal Swamp National Wildlife Refuge (GDSNWR). The GDSNWR was considered to be the dry site, while Alligator River National Wildlife Refuge (ARNWR) was set as the wet site. Data was collected by various students in Dr. Atkinson's research lab from 2005 to 2016. Atlantic White Cedar cores were mounted and sanded with progressively finer sandpaper until the tree rings were evident. Cores and cross sections were cross-dated and analyzed. Basal

area increment (BAI) was calculated to determine the total area of each tree ring. The mean annual BAI in the GDSNWR reference site was larger than the mean in the ARNWR reference site. All unknown site BAI averages were more similar to the GDSNWR reference site. These results may be used to evaluate site historic water levels which may assist in establishing water level targets and in facilitating AWC reestablishment.

Chemistry

PHOTOSWITCHABLE ARYLAZOPYRAZOLE LIGANDS: SYNTHESIS AND COORDINATION CHEM. Keset Y. Ghebreyessus and Janay I. Little, Department of Chemistry and Biochemistry Hampton University, Hampton, VA 23668. Novel palladium(II) and platinum(II) coordination complexes incorporating pyridine and benzothiazole functionalized photo-switchable arylazopyrazole ligands were synthesized and characterized. The photo-physical properties of the ligands and their corresponding metal complexes have been investigated. The effect of the nature of the ligands and type of metals on the rate of the *cis*-to-*trans* photo-isomerization process has also been explored. The ligands and the metal complexes exhibit strong absorption bands in the UV region, and weak bands in the visible region. Upon irradiation with UV-light ($\lambda = 365$ nm) these compounds clearly undergo efficient and almost quantitative *trans*-to-*cis* photo-isomerization of the azopyrazole unit followed by the reverse *cis*-to-*trans* isomerization upon standing under normal light. The results indicate that the percent conversion of the *trans*-to-*cis* isomerization is highly dependent on the substitution pattern on the pyrazole ring, which also has a significant influence on the electronic properties of the compounds. (Supported by: the Hampton University Faculty Research Fund, NSF CREST Center and PREM)

PROGRESS TOWARD A "GREEN" REDUCING AGENT. Charles M. Bump, Department of Chemistry and Biochemistry, Hampton University 23668. Solutions of freshly brewed green tea have been successfully used in the synthesis of gold and silver nanoparticles. During that process, the tea reduces metal ions (Au^{3+}, Ag^+) to their elemental state and caps those atoms with a large organic molecule that prevents the aggregation and precipitation of colloidal metal. Among the more conventional reducing agents used in the synthesis of nanoparticles is $NaBH_4$. That same reducing agent is widely used for the conversion of aldehydes and ketones to alcohols. We are beginning an investigation on expanding the use of green tea for the reduction of aldehydes and ketones. Some of the difficulties to be addressed include the poor solubility of ketones in aqueous media (e.g. tea), determining the length of time required for the reduction to take place, and the isolation of the reduction product.

MICROWAVE-ASSISTED SYNTHESIS AND CHARACTERIZATION OF METALLIC NANOMATERIALS Peter N. Njoki, Department of Chemistry & Biochemistry, Hampton University, Hampton VA 23668. Laboratory microwaves are becoming common tools in research and teaching laboratory. Compared to conventional means of heating, microwaves provide shorter reaction times, better reproducibility, and enhanced reaction control. The key component of microwave-assisted heating is that nanoparticles synthesis can be completed within minutes thereby saving time and energy. Nanomaterials possess novel properties as the size of particles decreases to nanoscale. To understand and exploit these novel properties, we need to develop effective strategies to synthesize nanoparticles with controllable size, shape, and composition. In this presentation, we focus on synthesis of gold and silver nanoparticles due to their unique optical

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properties and ease of fabrication. This presentation describes results of a microwave-assisted irradiation method aimed at developing the ability to control the size and composition of gold and silver nanoparticles. We will also discuss applications of these nanoparticles in fuel cell and forensic analysis. An array of analytical techniques such as transmission electron microscopy and UV-Vis spectrophotometry were used for the characterizations.
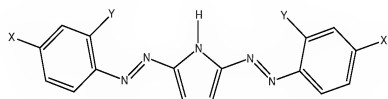
ENGAGING INQUIRY-BASED STUDENT LEARNING USING COMPUTATIONAL CHEMISTRY IPAD APPS IN ORGANIC CHEMISTRY LABORATORIES. Michelle K. Waddell, Charles Bump, Godson Nwokogu & Edmund Ndip, Dept. of Chem. and Biochem., Hampton Univ., Hampton VA. 23668. Inquiry-based classrooms encourage students to explore and understand their world using scientific methods. Spartan has been used to integrate computational chemistry into undergraduate chemistry curriculum. Computers and modeling software were purchased for use in chemistry courses. However, calculations using Gaussian and Argus Lab software to minimize energies of organic molecules routinely ran for several hours and are not feasible for use in the organic laboratory sections. The free website 4 Mol D was adopted for its quick efficient calculations. This project compares organic chemistry students' comprehension of inquiry-based learning of computational chemistry 3D models of various experiments using 4 Mol D and the iPad mini iSpartan app. Student outcomes were assessed in retrospective web-based surveys administered on-line through Blackboard $_{TM}$ 9.0 to access comprehension of the material, level of success in achieving learning outcomes and technical ability to utilize the molecular modeling applications. Results indicated that students were more comfortable using the iPad minis due to its ease of use and reliability of access. Student data collected from iPad minis and 4 Mol D was comparable to one another.

GC/MS QUANTIFICATION OF THE BIOFUEL POTENTIAL OF ALGAE. Grant A. McClure & Dr. Todd Allen, Dept. of Biol. and Chem., Liberty Univ., Lynchburg VA. 24502. Microalgae has the potential to be used as an alternative to petroleum-based feedstock necessary for transportation fuels. Several companies are researching cost-effective ways to optimize algae growth and harvesting techniques in an effort to convert the lipid fraction of the algae biomass into various types of fuels. The ability to accurately quantify the lipid content of algae is essential to evaluating the fuel potential. A validated GC/MSD (Gas Chromatography coupled with Mass Spectrometric Detection) method was previously developed for the quantitative analysis of the biofuel potential in algal samples provided in lyophilized and filtered mediums. Companies are exploring new oil extraction techniques to maximize the harvest efficiency of the oil from the algae. The new oil extract required additional steps be taken in the beginning stages of the sample preparation to accommodate the unique matrix. After the additional steps for the new matrix, all subsequent steps merge with the other matrices preparation method. Results will be presented.

MICROWAVE-ASSISTED SYNTHESIS AND CHARACTERIZATION OF METALLIC NANOMATERIALS Peter N. Njoki, Department of Chemistry & Biochemistry, Hampton University, Hampton VA 23668. Laboratory microwaves are becoming common tools in research and teaching laboratory. Compared to conventional means of heating, microwaves provide shorter reaction times, better reproducibility, and enhanced reaction control. The key component of microwave-assisted heating is that nanoparticles synthesis can be completed within minutes thereby saving time and energy. Nanomaterials possess novel properties as the size of particles decreases to nanoscale. To understand and exploit these novel properties, we need to develop

effective strategies to synthesize nanoparticles with controllable size, shape, and composition. In this presentation, we focus on synthesis of gold and silver nanoparticles due to their unique optical properties and ease of fabrication. This presentation describes results of a microwave-assisted irradiation method aimed at developing the ability to control the size and composition of gold and silver nanoparticles. We will also discuss applications of these nanoparticles in fuel cell and forensic analysis. An array of analytical techniques such as transmission electron microscopy and UV-Vis spectrophotometry were used for the characterizations.

EFFECTS OF SUBSTITUENTS AND BRIDGE LENGTH ON NLO PROPERTIES OF A SERIES OF BIS AZO DYES. Edmund Moses N. Ndip, David S. Barnes & Binal Patel. Department of Chemistry and Biochemistry, Hampton University, Hampton, VA 23668. In the present study, the effect of variations in donor – acceptor pairs on frequency dependent nonlinear optical properties has been investigated. Semi-empirical calculations of the frequency-dependent linear polarizability, α, the second, β- and third, γ-order hyperpolarizabilities at the fundamental (1028 nm) and other wavelengths were achieved by applying the *ab initio* time-dependent coupled perturbed Hartree -Fock (TDHF) method at the Restricted Hartree- Fock (RHF) level using routines contained in MOPAC2012 program for various model systems shown below (1-10). The D-π-D structures were found to have lower bandgaps (6.585 – 7.004 eV) as well as significantly lower second-order hyperpolarizability values compared to values for A-π-A, A-π-D, or D-π-A structures.

Compound #	X	X	Y	Y	Type
1	H	H	H	H	
2	CO_2H	CO_2H	H	H	A-π-A
3	NH_2	NH_2	H	H	D-π-D
4	CO_2H	NH_2	H	H	A-π-D
5	OH	OH	H	H	D-π-D
6	CO_2H	OH	H	H	A-π-D
7	NH_2	OH	H	H	D-π-D
8	H	H	OH	OH	No type
9	NPH_2	NPH_2	H	H	D-π-D
10	N-Carbazoyl	N-Carbazoyl	H	H	

CHEMISTY AND SPECTROSCOPY IN KBR MATRICES, T.C. DeVore, Dept of Chem. and Biochem., James Madison University, Harrisonburg VA 22807. Fifty years ago, several methods for trapping ions in KBr crystals were discovered. The IR spectrum of these ions could then be investigated and chemical reactions involving these ions could be investigated by heating the crystal. One way to use this method in the chemistry labs is to investigate the spectra of isoelectronic series of molecules. One example of this presented is the changes in the bond strengths for 16 electron linear molecules BO_2^-, CO_2, and NCO^-. The IR spectrum of BO_2^- shows a clear isotope effect and can also be used to measure the isotopic abundance of boron. A similar exercise can be done with the isoelectronic tetrahedral molecules/ ions BH_4^-, CH_4, and NH_4^+. The thermal decomposition of the oxalate ion in KBr produced by allowing an aqueous solution containing 1% sodium oxalate- 99% KBr to evaporate to dryness. Heating the solid to 775 K

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produced 4 new IR bands that may arise from $COCO_3^{2-}$ which has been predicted to be an intermediate in this decomposition.

Posters

CHARACTERIZATION OF ALGAL PIGMENTS USING ABSORBANCE AND FLUORESCENCE. C. M. Fleming, Z. J. Schreiber & T. M. Allen, Department of Biology and Chemistry, Liberty University, Lynchburg VA 24502. The effectiveness of identifying different types of chlorophyll within microalgae is vital in predicting not only the lipid content, but also understanding why chlorophyll is contaminating the hexane layer during the sample preparation process. This contamination of chlorophyll and other proteins inhibits the extracted lipids from immediately being used for biofuels. A comparison of absorbance and fluorescence in both the organic and aqueous layers were measured to analyze the ratio between extracted and non-extracted pigments found in the organic layer.

Data Science, Computing and Statistics

DISCOVERING RASPBERRY PI. Bruce C. Chittenden, Department of Computer Science, Hampton University, 100 E. Queen Street, Hampton, Virginia 23668. The Raspberry Pi is a low cost, credit-card sized single-board computer that plugs into a HDMI monitor and uses a standard USB keyboard and mouse. This paper outlines how the Raspberry Pi was introduced and incorporated into CSC 301 - Operating Systems at Hampton University. A series of program assignments were developed utilizing the Raspberry Pi to enhance students' learning in Operating Systems concepts. The feedback from the students was incredibly positive as they really enjoyed the hands-on nature of working with the Raspberry Pi and the actual source code for the Linux Operating System.

BIG DATA CONTENT MINING TECHNIQUES FOR DETECTING CYBERBULLYING IN SOCIAL MEDIA. Yen-Hung Hu & Shawniece L. Parker, Dept. of Computer Sci., Norfolk State Univ., Norfolk, VA 23504. Social media have become an increasingly popular trend and teenagers have made up a large percentage of their users. Teens often disregard online safety rules and guidelines and are more inclined to misuse social media because they are often unaware of the privacy rights associated with the use of that particular media. As a result, cyberbullying cases have steadily risen in recent years and some have gone undiscovered, or are not discovered until serious harm has been caused to the victims. This study aims to create an effective algorithm that can be used to detect cyberbullying in social media using content mining. Because bullies may not use only one social media to victimize other users, the proposed algorithm must detect whether or not a user is victimizing someone through one or more social media accounts and determine which social media accounts are being used to carry out the victimization. To achieve this goal, the algorithm will collect information from contents shared by the victims in all of their social media accounts, then will determine which contents to extract, based on a big data technology, those pre-defined phrases or words that might be used by cyberbullies. Any extracted data will reveal some insight into whether or not cyberbullying is occurring and trigger appropriate approaches to handle it.

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REVIEW OF SECURED MOBILE APPLICATION DEVELOPMENT PROCESS IN THE INDUSTRY. <u>Yen-Hung Hu</u> & Thomas K. Annan, Dept. of Computer Sci., Norfolk State Univ., Norfolk, VA 23504. Mobile applications use has heavily increased in recent times and this upsurge comes with certain security concerns which lead to potential security threats to the developers as well as the end users. These concerns could be minimized and controlled if an appropriate secure software development process has been suggested and implemented. However, such a suggestion is not always being adopted due to a limit budget and timeline. In this research we study potential mobile application vulnerabilities, review agile software development process, and investigate the integration of security implementations into agile software development process. The research also conducts a case study to review how a software company applies secured application processes in their mobile apps development irrespective of adopted process models. Based on the case study, recommendations of how to integrate required security components into agile software development process were assessed and suggested.

Education

A QUANTITATIVE ANALYSIS OF VIRGINIA PUBLIC SCHOOL SPECIAL EDUCATION DUE PROCESS CASES AND THEIR RESOLUTIONS FROM 2004-2016. <u>Kurt Y. Michael</u>, Deanna L. Keith & Valerie A. Beaudoin-Saunders, School of Education, Liberty University, Lynchburg, VA, 24515. This study analyzed data from the Virginia Department of Education's due process database over a 12-year period of time and noted common disabilities involved in special education litigation. A descriptive content analysis was used to examine the total frequencies regarding types of disability cases and their outcomes. Data was obtained from the Virginia Department of Education's Office of Dispute Resolution and Administrative Services. The types of disabilities examined came from the thirteen categories of special education as defined by the Individuals with Disabilities Education Act. The data collected was in the form of frequency counts that were then analyzed using tables and trend charts.

FRAMING SUSTAINABILITY IN THE FIELD: REFLECTIONS OF AN IMMERSIVE EXPERIENTIAL (E-TERM) COURSE IN VIRGINIA AGRICULTURE. Tim C. Durham, Agriculture Program, Ferrum College, Ferrum, VA 24088. Often ill-defined and difficult to quantify, sustainability is implicitly at the nexus of all agricultural activities. To date, there have been myriad courses that examine the theories underpinning sustainability, but a dearth of courses that contextualize the seemingly unsolvable equation from a multifaceted, field perspective. Recognizing the crossover appeal that such a course could provide at a small liberal arts institution, AGS 218: Regional Experiences in Agriculture was developed for an inaugural offering in 2014, and offered again in 2016. Over three immersive weeks, participants examined agriculture in its many incarnations, including commodity groups, avant-garde systems including agroecology and hyperyielders, on-farm alternative energy generation, precision ag, HBCU's, policy development and advocacy in Washington, D.C., farmland preservation, urban edge agriculture, green roofs, marketing, eco-consumerism, food miles, eco-justice, and the intersections between Cooperative Extension, Land Grant institutions, and Agricultural Experiment Stations. Students were challenged to evaluate the merits of different operations within the three established pillars of sustainability. They were reminded of the often tenuous intersections between agriculture and natural ecologies, and the incumbency and duty of farmers as stewards. Moreover, students learned that sustainability is site-specific and not a one size fits all proposition. As a result, participants are

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better positioned to make meaningful contributions to this mission critical landscape as change agents in their respective fields.

DEVELOPING A MENTORING NETWORK ACROSS VIRGINIA FOR TRAINING IN ACTIVE LEARNING TECHNIQUES. Michael J. Wolyniak, Dept. of Biology, Hampden-Sydney Coll., Hampden-Sydney VA 23943. The undergraduate science education community has responded to the recommendations of the American Association for the Advancement of Science (AAAS) *Vision and Change* document with several initiatives designed to improve the way in which undergraduates learn science. These initiatives have often taken the form of one-time workshops that generate awareness of and interest in active learning techniques among participants. However, they have been less successful with respect to generating the sustainable change necessary to bring real reform to undergraduate science education. To create sustainable change, long-term faculty development initiatives focused on mentorship are needed so that instructors seasoned in active learning can convey their experiences to mentees interested in using these pedagogical techniques as the centerpiece of their own teaching. The Virginia Academy of Science (VAS) is well positioned to be a leader in providing current and future instructors with the training necessary to effectively bring authentic research into their classrooms. Here, I discuss the efforts of other professional societies like The American Society for Cell Biology (ASCB) to develop a mentorship network for the spread of best practices in teaching and how the VAS can adapt this model to become a leader in the development and spread of instructors equipped to utilize course-based research experiences (CUREs) in their teaching.

TARGETED INFUSION PROPOSAL: VIRGINIA UNION UNIVERSITY RESEARCH TRAINING PROGRAM IN THE BIOLOGICAL SCIENCES-YEAR ONE. Carleitta Paige-Anderson, PhD, Center for Undergraduate Research and Department of Natural Sciences, Virginia Union University, Richmond, VA 23220. Targeted Infusion Projects awarded through Historically Black Colleges and Universities Undergraduate Program (HBCU-UP) support the development, implementation, and study of innovative evidence-based approaches to improve the preparation and success of HBCU undergraduate students pursuing STEM graduate programs and/or careers. This project focuses on the Biological Sciences major at Virginia Union University (VUU) and seeks to address interrelated issues facing STEM departments at many small, liberal-arts institutions, such as: 1) limited access to meaningful research experiences for undergraduates and 2) obstacles to faculty research productivity. To address these issues, the project will utilize a comprehensive approach to integrate research into the curriculum and bolster faculty research capacity. The project will incorporate course-based undergraduate research experiences into introductory Biology courses, provide Biology majors access to scientific research-related co-curricular activities and resources, and establish a Biology capstone research project. The faculty research interests will serve as a basis for these authentic research activities. It is anticipated that this project will enhance the quality of STEM education at VUU, leading to the formation of a new research-intensive academic curriculum, and increase VUU's production of STEM graduates who are competitive for graduate school and careers in biological sciences. (Supported by: National Science Foundation Award: 1623357).

CLIMATE CONNECTIONS IN VIRGINIA: YOUR ACTIONS MATTER. Jeremy S. Hoffman, Eugene G. Maurakis and Richard C. Conti, Science Museum of Virginia, Richmond, VA 23220. Our project objectives are to communicate the science of climate change on global and local scales,

highlight current and potential future impacts of climate change on Virginia and its communities, define community climate resiliency and why it is important, illustrate how individuals can contribute to the resiliency of their own community by taking personal steps to be prepared for weather events and health threats related to climate change, and foster a conversion of climate change awareness and understanding into personal action to increase readiness and resiliency in homes, schools, and communities. Climate change and resiliency information were conveyed by the development of new programming for the museum's NOAA Science on a Sphere® and digital Dome theater, production of a statewide digital media series (24 audio and 12 video content pieces/year), engagement with social media platforms, a public lecture series, facilitation of resiliency-themed programming (Art Lab, Challenge Lab, EcoLab), establishment of extreme event readiness challenge workshops, and enacting community preparedness and resiliency checklist and certification programs. Formative evaluation indicated that around two-thirds of survey respondents liked the visual display of the Science On a Sphere; about half of respondents enjoyed working on teams during the Extreme Event Challenge, and a number of respondents enjoyed the content presented in our Dome show, "The Cosmic Climate Cookbook." Points of confusion between programs and within programs identified by respondents also are being addressed. (Funded by NOAA Award NA15SEC0080009, Virginia Environmental Endowment, and Virginia Sea Grant).

CREATIVE CHANGE: ART, MUSIC, AND CLIMATE SCIENCE. Rebecca A. Dahlberg & Eugene G. Maurakis, Science Museum of Virginia, Richmond, VA 23220. As the effects of climate change grow, so too does the need for effective climate science education and engagement to increase public resiliency. Studies at Columbia University suggest traditional approaches, relying heavily on scientific information and data, fail to engage and inspire action in most people. Multidisciplinary approaches are key to improve the next level for climate change communication to stimulate behavioral change. Science Museum of Virginia hosted *Creative Change* in March 2017, a multidisciplinary event reacting to and interpreting climate change and resiliency through a variety of artistic mediums, including music, dance, visual arts, theater, and poetry. Our goal was to increase public awareness of climate change and resiliency through the arts and humanities in support of the Museum's mission to *inspire Virginians to enrich their lives through science*. Featured programming included *Coral Reef Fever*, a dance performance of coral bleaching; high school and university art exhibitions; climate data performed by a string quartet; poetry, rap, and theater performances; and a panel discussion by artists and scientists on communicating science through the arts and humanities. To evaluate the effectiveness of the programming, 26 Museum visitors completed surveys. Results indicated that visitors enjoyed the event (mean of 9.58 out of 10), learned new information (9.07), and strongly agreed that the arts and humanities should be used more in communicating science concepts (9.77). (Funded in part by Bond Bradley Endowment and NOAA ELG Award #NA15SEC0080009).

INTERPRETING SCIENCE THROUGH ARTS AND HUMANITIES. Eugene G. Maurakis, Biology Dept., University of Richmond, VA 23173 and Science Museum of Virginia, 2500 W. Broad St., Richmond, VA 23220. Objectives were to provide learning opportunities and platforms for students to express science phenomena through the arts and humanities, increase student engagement, and enhance student appreciation of multiple viewpoints. Hypotheses were that arts and humanities do not give students a different perspective of science, and that students do not find arts and humanities valuable in communicating science. Seven undergraduates and one

graduate student (represented by three females and five males) comprised the non-major liberal arts biology course composed of lectures in science, ecological principles, evolution, and ethics; discussion assignments of peer-reviewed articles; in-class guest lecturers in music, art, theater, and poetry; and out-of-class lectures by seven scientists on climate change and its impacts on Virginia. Each student selected a specific environmental issue, researched ecological impacts, and created an artistic expression of their topic, which was displayed in an on-campus exhibition. On a scale of one (highly disagree) to 10 (highly agree), survey results indicated: students learned new information from the professor's lectures (9.8), guest lectures (9.8), and off-campus scientists (9.8), learned new information about how the arts and humanities communicate science (10), arts and humanities communicate science in ways that science cannot (10), arts should be used more in communicating science (10), and students are interested to take other such multidisciplinary courses (10). Funded in part by University of Richmond's Bonner Center for Civic Engagement.

EVALUATING LIGHTPLACE: AN EARLY CHILDHOOD EXPERIENCE. Jessica E. Scalise[1], Rebecca A. Dahlberg[2], Eugene G. Maurakis[2] & Gary N. Crayton Jr.[2], [1] SUNY Oneonta, Cooperstown Graduate Program, Cooperstown, NY, 13326 and Science Museum of Virginia, Richmond, VA, 23220. Science Museum of Virginia opened LightPlace in Spring, 2016, an early childhood experience space of 18 interactive platforms, each with multiple activities specifically designed for children ages 0-5. LightPlace is divided into two areas: one for toddlers and preschoolers (2-5 years) and the other for infants (0-18 months). The primary 18 activities in the toddler-preschool area focus on STEM and early childhood developmental skills relative to Virginia Foundation Blocks for Early Learning. Museum staff and interns conducted an evaluative study to observe children's interactions with activities in LightPlace, survey caregivers, and test the effect of suggestion cards at seven activities. A total of 86 observations were made: 42 observations of children and caregivers, and surveys of caregivers prior to the introduction of interaction suggestion cards; and 44 observations and surveys after the introduction of the interaction suggestion cards. We found museum guests spend an average of 31.2 minutes in LightPlace and rated it at 4.81 out of 5 for satisfaction. Additionally, suggestion cards did not significantly change the level of initial caregiver interaction with children (94%; p=0.0005), but caregivers gave twice as many demonstrations (70.4%) with the suggestion cards. Many parents said they did not know about LightPlace but commented that it was the main reason they decided to get a membership to the Museum when they discovered it.)Funded by the Nunnally Foundation, Memorial Foundation for Children, and The Community Foundation).

Entomology

THE ANTS OF VIRGINIA PROJECT. Kal Ivanov, J. B. Keiper, & L. Hightower, Department of Recent Invertebrates, Virginia Museum of Natural History, Martinsville VA, 24112. No comprehensive treatment of the ants of Virginia exists with the majority of the published species accounts found scattered throughout the literature. The Ants of Virginia Project (AVA) was initiated in late 2014 when a thorough review of the published literature revealed that a total of 122 ant species were known to occur in the state. These numbers represent a stark underestimation of the true number of species expected to occur in the state given the diversity of ecoregions and habitats found in Virginia, and especially when viewed in the light of recent updates to the ant lists of some neighboring regions. A systematic state-wide survey, coupled with visits to museum collections and a review of available online databases, was initiated in the spring of 2015. As a

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result of our efforts additional 28 records are added to the list. As of May 2017 a total of 150 species, including 12 ant exotics, are reliably reported from the state. These species represent most of the North American groups, lacking only the more tropical members of Ectatomminae and Pseudomyrmecinae. The most speciose genera in the state are S*trumigenys* (19 species), *Formica* (18), *Lasius* (11), and *Aphaenogaster* (10). With this project we aim to bridge the gap between our current understanding of the state's ant diversity and distribution and the true diversity of Virginia's myrmecofauna where new discoveries are undoubtedly to be expected.

FACTORS AFFECTING NEST SELECTION BY *TEMNOTHORAX* SPP. ANTS (FORMICIDAE). Chelsea Hughes & Deborah Waller, Dept. of Biology, Old Dominion University, Norfolk, VA 23529. *Temnothorax*, commonly called acorn ants, nest inside hollowed twigs or acorns and have been shown to consider and weigh several variables such as size and shape when choosing the most desirable nest. *T. curvispinosus* nest preferences have been studied and documented previously, but no research has yet been published regarding choices and behavior of *T. tuscaloosae*. This study compares the behavior of the two species over the course of two experiments, each comparing different parameters of nest sizes and shapes. Experiment one focused on four sizes of acorn-shaped artificial nests. *T. curvispinosus* made choices consistent with findings of previous research, but *T. tuscaloosae* preferred nesting in the very small entrance tunnels only. Experiment two compared twig-shaped nests with an acorn-shaped nest. *T. curvispinosus* preferred the smallest tube nest in this case and *T. tuscaloosae* preference was varied. The results from this study indicate that preference of twig-shaped nests may be stronger for *T. curvispinosus* than previously thought, and that many unestablished variables appear to drive the decision-making process for *T. tuscaloosae.*

MOLECULAR INSIGHTS INTO VECTOR-PATHOGEN-HOST INTERACTIONS. Girish Neelakanta, Center for Molecular Medicine, Department of Biological Sciences, Old Dominion University, Norfolk, VA 23529. In the United States, hard ticks *Ixodes scapularis* and soft ticks *Ornithodoros turicata* transmits various pathogens to humans that include *Anaplasma phagocytophilum*, the agent of Human anaplasmosis and *Borrelia turicatae*, the agent of human relapsing fever, respectively. These ticks ingest pathogens upon feeding on an infected animal. Hard and soft ticks show considerable differences in the developmental life cycle stages, blood feeding and reproductive behaviors. The mechanism(s) at the molecular level that can explain the major differences in these key biological processes between these ticks are not known. In the view for the development of an anti-vector vaccine as an effective means to block transmission of several pathogens from these ticks, understanding the role of conserved arthropod molecules in vector biology and interactions with pathogens remains important. In this study, several molecular findings in understanding vector biology, pathogen survival in the athropod cells and development of transmission-blocking strategies to interrupt live cycle of these ticks are discussed.

NOVEL TRANSMISSION BLOCKING STRATEGIES USING ARTHROPOD EXOSOMES. Hameeda Sultana, Center for Molecular Medicine, Department of Biological Sciences, Old Dominion University, Norfolk, VA 23529. Molecular determinants and mechanisms of arthropod-borne flavivirus transmission to the vertebrate host are poorly understood. The transmission strategies used by flaviviruses to exit arthropods and infect human host were envisioned as best approaches to develop transmission-blocking vaccines against vector molecules or determinants that facilitate pathogen transmission. Research in my laboratory has shown that both tick and

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mosquito-borne flaviviruses uses exosomes, the small membranous extracellular vesicles for transmission from arthropods to human host. Our studies have revealed that arthropod derived exosomes are important means of communication and transmission between the vector and the vertebrate host. We have found that Langat virus (LGTV), a flavivirus member closely related to tick-borne encephalitis virus and mosquito-borne dengue/ZIKA viruses are transmitted from vector to the vertebrate host through extracellular vesicles or exosomes. The exosomes containing flaviviruses viral RNA and proteins were stable, secured and highly infectious in all tests such as re-infection kinetics, trans-migration and viral plaque formation assays suggesting these extracellular vesicles as favorable modes of transmission. Taken together, exosomes serves as protective vesicles for transmission of vector-borne flaviviruses that escape the vertebrate immune system and cause pathogenesis and perhaps death. Our current efforts are focused on understanding the molecular mechanisms and associated signaling cascades involved in mediating the arthropod derived extracellular vesicles that facilitate pathogen transmission to the vertebrate host.

DETECTABLE TRACES OF MERCURY [THg] EXIST WITHIN INVERTEBRATE FOOD WEBS OF LOW CONTAMINATION SHENANDOAH VALLEY TERRESTRIAL ECOSYSTEMS. <u>Melissa E. Encinias</u>, Gregory E. Mansour, Matthew W. Riordan, Cody J. Cubbage & W. Dean Cocking, Dept. of Biol., James Madison Univ., Harrisonburg, VA 22807. Inorganic and organic background Mercury (Hg), have been increasing in the environment. This can be attributed to both natural and anthropogenic sources and is known to negatively impact biological systems. Airborne Hg pollution puts at risk ecosystems previously thought to be uncontaminated. To identify the possible long term implications of Hg accumulation in invertebrates inhabiting "uncontaminated" terrestrial environments, four successional forest sites in and around Rockingham Co. VA, were sampled over the summers of 2014, 2015, and 2016. Replicate habitat and invertebrate samples were analyzed for THg through Atomic Absorption Spectrophotometry using a Flow Injection Mercury System (FIMS). This analysis revealed detectable trace THg concentrations in soil, duff and tissues of representative invertebrates. The study supports the prediction of greater THg concentrations being associated with invertebrates occupying higher trophic levels, even at low contamination levels. Invertebrates, such as beetles and earthworms show potential as trace level bio-monitors within sites that would be low concentration controls in a study of high level THg contamination.

COORDINATION OF TWO THIOSEMICARBAZONES TO COPPER(II) AND COBALT(III) METAL CENTERS ENHANCES ZIKA VIRUS AND DENGUE VIRUS LOADS IN BOTH ARTHROPOD CELLS AND HUMAN KERATINOCYTES. <u>Shovan Dutta</u>[1], Alexis Huddleston[2,#], Supreet Khanal[1], Colin Simms[1], Jessa Faye Arca[2,#], Amlan Mitra[2,#], Loree Heller[3,4], Piotr J. Kraj[1,5], Michel Ledizet[6], John F. Anderson[7], Girish Neelakanta[1,5], Alvin A. Holder[2] & Hameeda Sultana[1,5*]. [1]Dept. of Biological Sciences, [2]Dept. of Chem. & Biochem, [3]Frank Reidy Research Center for Bioelectrics, [4]School of Medical Diagnostic & Translational Sciences, [5]Center for Molecular Medicine, ODU, Norfolk, VA, [6]L2 Diagnostics LLC, [7]Connecticut Agricultural Experiment Station, CT. Copper and cobalt have been associated with virus-host interactions. We show that four compounds 2-acetylethiazole (compound 1), 2-acetylthiazole thiosemicarbazone ligand-copper(II) complex (compound 2), thiosemicarbazone ligand-derived from citral (compound 3) and citral-thiosemicarbazone ligand-cobalt(III) complex (compound 4) increased DENV2 and ZIKV replication in both C6/36 and HaCaT cells. Treatment

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with compounds 2 or 4 enhanced ZIKV and DENV2 transcripts in HeLa and mouse bone marrow derived dendritic cells. Also, pre- or post-treatments with conjugated compound 2 or 4 showed higher loads of ZIKV or DENV2 in HaCaT cells. These findings suggest that use of Cu(II) or Co(III) conjugation could enhance DENV2/ZIKV loads in human cells and elevate pathogenesis in infected individuals or individuals pre-exposed to such conjugated complexes. (Supported by: ODU startup funds to HS).

INVESTIGATING HETEROGENEITY OF *EHRLICHIA CHAFFEENSIS* PREVALENCE: MULTI-YEAR DATASET REVEALS INFLUENCE OF SEASONAL WEATHER PATTERNS. Dylan T. Simpson[1], Matthias Leu[1], Molly Teague[1], Johanna Weeks[1], Oliver Kerscher[1], Phillip D'Addio[1], Joseph Thompson[1], Alan Harris[1], Andrew Lewis[1], Richard Canella[1], Julia Moore[2], Brent Kaup[2], [1]Dept. of Biol., [2]Dept. of Sociology, College of William and Mary, Williamsburg, VA, 23185. *Ehrlichia chaffeensis* is a pathogenic bacterium transmitted by the lone star tick (*Amblyomma americanum*) and maintained by the white-tailed deer (*Odocoileus virginianus*) as its primary reservoir. Incidence of human infection by *E. chaffeensis* is increasing in the US, but its dynamics remain poorly understood. During June and July of 2012, 2013, 2015, and 2016, we studied dynamics of *E. chaffeensis*, its hosts, and environmental factors at 130 sites across the Virginia Peninsula. We collected lone star ticks and used PCR to test for *E. chaffeensis.* Using generalized linear mixed-effect models, we asked what factors drive inter-annual variation in *E. chaffeensis* prevalence and tick abundance. Most importantly, we found that prevalence is positively associated with rainfall during the current spring, and with temperature the previous fall and winter, while tick abundance is associated with precipitation during the current spring and temperature the previous spring. We hypothesize precipitation to affect tick survival through the relief of water stress, and temperature to affect deer and tick fecundity. Our results highlight the complexity of tick-borne disease dynamics, but also represent an important step toward modeling *E. chaffeensis* prevalence through time.

IXODES SCAPULARIS ORGANIC ANION TRANSPORTER IS REQUIRED FOR *ANAPLASMA PHAGOCYTOPHILUM* SURVIVAL AND TRANSMISSION FROM TICKS. Vikas Taank[1], Shovan Dutta[1], Amrita Dasgupta[1], Tanner K Steeves[2], Durland Fish[2], Ulrike Munderloh[3], John F Anderson[4], Hameeda Sultana[1,5], & Girish Neelakanta[1,5]. [1]Department of Biological Sciences, ODU, Norfolk, VA, [2]School of Public Health, Yale University, New Haven, CT. [3]Department of Entomology, University of Minnesota, St Paul, MN, [4]Department of Entomology, Connecticut Agricultural Experiment Station, New Haven, CT. [5]Center for Molecular Medicine, College of Sciences, ODU, Norfolk, VA. *Ixodes scapularis* ticks transmit the human anaplasmosis agent, *Anaplasma phagocytophilum*. In this study, we show that *A. phagocytophilum* specifically up-regulates *I. scapularis* organic anion transporting polypeptide, isoatp4056 and kynurenine amino transferase (kat), a gene involved in the tryptophan metabolism, for its survival in ticks. RNAi analysis revealed that knockdown of isoatp4056 or kat or both affected *A. phagocytophilum* survival in tick cells in vitro. Exogenous addition of xanthurenic acid (XA), a metabolite from tryptophan pathway, induces isoatp4056 expression and *A. phagocytophilum* burden in ticks and tick cells. EMSA assays provide further evidence that *A. phagocytophilum* and XA influences isoatp4056 expression. Collectively, this study serves as a model to understand the interplay between molecular pathways manipulated by a pathogen to survive in its arthropod vector. (Supported by: ODU Start-up funds to GN and HS).

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TICKS ELICIT VARIABLE FIBRINOGENOLYTIC ACTIVITIES UPON FEEDING ON HOSTS WITH DIFFERENT IMMUNE BACKGROUNDS. <u>Ashish Vora</u>[1], Vikas Taank[1], Sucharita M. Dutta[2], John F. Anderson[3], Durland Fish[4], Daniel E. Sonenshine[1], John D. Catravas[5,6], Hameeda Sultana[1,7] & Girish Neelakanta[1,7], [1]Department of Biological Sciences, ODU, VA, USA. [2]Leroy T. Canoles Jr. Cancer Research Center, EVMS, VA, USA. [3]Department of Entomology, Connecticut Agricultural Experiment Station, New Haven, CT, USA. [4]School of Public Health, Yale University, New Haven, CT, USA. [5]Frank Reidy Research Center for Bioelectrics, ODU, VA, USA. [6]School of Medical Diagnostic and Translational Sciences, College of Health Sciences, ODU VA, USA. [7]Center for Molecular Medicine, College of Sciences, ODU, VA, USA. Ticks secrete several factors in their saliva to suppress the host defenses against infestations. Using *I. scapularis* and mice from two different immune backgrounds as a model, we show that ticks fed on immunodeficient mice demonstrate decreased fibrinogenolysis in comparison to ticks fed on immunocompetent mice. Reduced levels of D-dimer and increased engorgement weights were evident in ticks fed on immunodeficient mice. The LC-MS/MS and QPCR analysis followed by inhibitor and antibody-blocking assays revealed that the arthropod HSP70-like molecule contributes to differential fibrinogenolysis. Collectively, these results indicate that ticks elicit variable fibrinogenolysis upon feeding on hosts with different immune backgrounds (Support: ODU startup funds for GN and HS.).

STAGE-SPECIFIC THERMAL LIMITS FOR GROWTH AND SURVIVAL IN AN INVASIVE FOREST PEST. <u>Nana Banahene</u>[1], Salem Salem[1], Hannah Byrne[2], Madison Glackin[2], Lily Thompson[1], Trevor Faske[2],Salvatore Agosta[2], Andrew Eckert[2] & Kristine Grayson[1], [1]Department of Biology, University of Richmond, Richmond, VA 23173 and [2]Department of Biology, Virginia Commonwealth University, Richmond, VA 23284. The spread of the gypsy moth (*Lymantria dispar* L.) across wide climatic gradients in North America provides an ideal system for studying the role of thermal limits in invasion. Previous work has shown that spread rate variability at the southern invasion front is correlated with supraoptimal temperatures. Gypsy moth individuals were exposed to daily temperature ramping cycles of either optimal (22-28°C) or supraoptimal temperature treatments (30-36°C, 32-38°C, or 34-40°C) at specific developmental stages (first through fourth larval instar, pupae) for either 2 or 7 days. We measured survival over the duration of the heat exposure and long term effects on development time and pupal mass. Survival generally decreased as treatment temperature increased for all stages. The 34-40°C temperature treatment had the largest effect on larval survival, with 7 days of exposure being lethal for all stages except second instar and pupae. All other temperature treatments had more than 75% survival after 7 days for all stages measured. The long term effects on final pupal mass of the 2 day exposure to heat were more pronounced for females than males and exposure to heat at later larval stages resulted in larger decreases in pupal mass. These results show negative effects of high temperature on gypsy moth populations and support patterns of spread seen at the southern invasion front.

EXPLORING HOKIE BUGFEST AND THE ENTOMOLOGY EVENTS IT PARALLELS. Stephanie L. Blevins, Department of Entomology, Virginia Polytechnic Institute and State University, Blacksburg, VA 24061. Hokie BugFest is an annual free event designed by the entomology department at Virginia Tech to interface with the public. Since its inception in 2011, the event has grown from 2,000 attendees to over 7,000 attendees in 2016. Entomology faculty, staff, and graduate students partner with Virginia Cooperative Extension, Virginia 4-H, and other entities to provide an educational experience to the public. The goal of hosting this outreach event

is to showcase research, increase public awareness and appreciation of entomology, and develop better public perceptions of insects and other arthropods. Although many universities host large entomology outreach events like Hokie BugFest, little research has been conducted to investigate these events and their impacts. Through literature review and web searches a sizeable list of institutions hosting large entomology outreach events were identified. Event coordinators at these institutions were contacted and asked to complete a Qualtrics survey pertinent to their event. The survey received an overwhelming response rate of 93%. Results revealed event information such as attendance, event structure, funding sources, popular exhibits, and impacts the event has on attendees, the hosting institution and the surrounding communities.

IDENTIFICATION OF MICROBIOTIA ASSOCIATED WITH THE ECTOSYMBIOTIC COMMUNITY STRUCTURE OF THE APPALACHIAN BROOK CRAYFISH (*CAMBARUS BARTONII*). Matthew M. Cooke, Luke Fischer, Sherrie Jeffers, Taylor Griffin & Kyle J. Harris. Department of Biology and Chemistry, Liberty University, Lynchburg, VA 24515. Within freshwater ecosystems, segmented worms (branchiobdellidans) exhibit a cleaning symbiosis with the Appalachian Brook Crayfish (*Cambarus bartonii*). Branchiobdellidans (e.g. *C. ingens*) have been shown to consume bacteria in the gill chambers and carapace of crayfish in addition to completing their life cycle on crayfish. However, little is known about the microbial community assembly in relation to crayfish and branchiobdellidans. This project explores the effects of branchiobdellidans on crayfish host microbial community assembly. Crayfish with and without branchiobdellidans were swabbed for bacteria after an eight week trial from individual study tanks. Isolated bacteria colonies that effectively grew on Trypticacse soy agar (TSA) had DNA extracted followed by PCR of the 16S gene. Sequencing was conducted by Eurofin Genomics and obtained sequences were placed in the Basic Local Alignment Search Tool (BLAST) for identification of genera. BLAST results confirmed six unique types of bacterial genera (*Shinella sp.*, *Aquitalea sp.*, *Agromyces sp.*, *Pseudomonas sp.*, *Pseudoxanthomonas sp.*, and *Acinetobacter sp.*) with no overlap with the worm treatment genera. Symbiont treatments confirmed four other types of bacterial genera (*Aeromonas sp.*, *Raoultella sp.*, *Bacillus sp.*, and *Micrococcus sp.*) with no overlap with the control genera. These initial findings suggest that crayfish carapace microbial assembly may change substantially with the presence or absence of branchiobdellidans.

Environmental Science

CHARACTERIZING 30 YEARS OF TREE, MOLD, AND GRASS POLLEN DATA FROM RICHMOND, VA. Rebecca Collie[1], Jeremy S. Hoffman[2], E.G. Maurakis[2] & J.M. Scanlon[2], [1]Allergy Partners of Richmond 23229 and [2]Science Museum of Virginia, Richmond, VA 23220. Seasonal pollen allergy symptoms comprise a significant amount of chief complaints in emergency department and urgent care center visits in the City of Richmond and across the Commonwealth of Virginia. As such, understanding long-term trends in seasonal pollen amounts and pollen season length would promote public health awareness campaigns aimed at improving readiness and resilience to longer and/or more intense allergy seasons. As seasonal pollen emissions reflect flowering phenology and productivity of regional pollen-producers, there is a strong sensitivity of flowering phenology to seasonal temperatures. Previous studies have shown modified pollen distribution patterns and phenological responses across space, time, and species. Here we examine the long-term and interannual trends in pollen seasons and pollen season durations across four types of pollen counted in the City of Richmond (tree, grass, ragweed, and mold) and seek to

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understand what, if any, environmental factors (e.g., frost-free season duration, mean annual minimum temperature, accumulated cyclone energy) follow similar trends. Initial results reveal several significant (p <0.05) linear trends over the 29-year record: 1) the date of peak counts of grass and tree pollen have moved earlier in the year, 2) the peak tree pollen load has increased, 3) the duration of the grass season has increased, 4) the ragweed season has gotten shorter and peak ragweed pollen load has decreased, and 5) the peak mold count has increased. We compare these trends with possible environmental forcing mechanisms and share plans for future work on these data.

INLET WIDTH CHANGE TRENDS FROM 1999 TO 2014 - VIRGINIA BARRIER ISLAND INLETS, VA. James D. Haluska, Department of Ocean, Earth, and Atmospheric Sciences, Center for Coastal Physical Oceanography, Old Dominion University, Norfolk, VA 23529. A total of 84 width measurements were made from 1999 to 2014 for each of the ten Virginia barrier island inlets was measured. Individual inlet changes showed that all inlets had substantial width changes during the analysis period. Great Machipongo Inlet had a net inlet width change of more than 1.9 km from 0.8 km in 1999 to 2.7 km in 2014. This was the largest width change of any of the ten inlets. Rapid inlet width changes for seven inlets occurred during four separate El Nino events. Data for the ten inlets was combined into a single data set and analyzed using EOF (PCA) analysis. The first EOF explained 95% of the data variance. Eigenvector 1, the spatial component of the PCA analysis, has a value of 0.95 corresponding to the Great Machipongo Inlet location. This was due to the extent of the change in this inlet versus the other 9 inlets. PC1 had a coefficient of correlation (r^2) to sea level rise of 0.77 and to wave height of 0.05. Continued inlet widening appears inevitable and will increase the risk of wave damage to areas to the west of the inlets.

A WATER QUALITY SUMMARY FOR CEDAR CREEK: SHENANDOAH, FREDERICK, & WARREN COUNTIES, VIRGINIA. Stephen M. Storrs & Woodward S. Bousquet, Environmental Studies Program, Shenandoah University, Winchester, VA 22601. Cedar Creek is an ecologically important 40 mile-long stream within the Shenandoah River watershed in northwestern Virginia. It flows into the North Fork of the Shenandoah River, from which a majority of the northern Shenandoah Valley communities obtain their drinking water. This meta-study's aim was to synthesize selected Cedar Creek water quality data to summarize the creek's overall water quality. We examined chemical and physical data from Friends of the Shenandoah River (FOSR), biological data from Shenandoah University based on VA Save Our Streams (VASOS) methods, and on EPA-DEQ (VSCI) methods. Chemical data from the FOSR's EPA- and DEQ-certified lab were nitrate nitrogen, orthophosphorous, ammonia, and pH. From 1997-present, each chemical factor displayed a high (79-100%) proportion of unimpaired readings. Turbidity was unimpaired 69.9% of the time. For 2014-present, 82.1% of the multi-metric VASOS scores for biological data fell into the acceptable range. The 2010 VSCI study, based on invertebrates keyed to family, revealed good to excellent water quality with the exception of a tributary stream flowing through agricultural land. The Virginia Department of Environmental Quality (DEQ) has not yet produced a TMDL (total maximum daily load) implementation plan for any potential water quality impairments. Taken together, the data examined reveal that Cedar Creek possesses clean and healthy water quality.

WATER QUALITY ASSESSMENT USING BENTHIC MACROINVERTEBRATES AND COLIFORM BACTERIA IN THREE MONTANE STREAMS IN CLARKE COUNTY,

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VIRGINIA. Courtney Bridges, Stephen Storrs, Woodward Bousquet, Nichole Davila-Sanchez, Madison Mackintosh, Andrew Richards, Munir Sayegh, Theresa Schottler & Alicia Sulc, Environmental Studies Program, Shenandoah University, Winchester, VA 22601. In 2013, Shenandoah University acquired its River Campus at Cool Spring Battlefield. The 195-acre property is situated along the Shenandoah River in Clarke County, Virginia. Our study examined three montane streams on the property in order to characterize the quality of water they contribute to the Shenandoah River. In keeping with U.S. EPA standards, the three tributary streams--Pigeon Hollow Run, Raven Rocks Run, and Rocky Branch--were each sampled in June and October 2016. Water quality assessments were made using two methods: the Virginia Stream Condition Index (VSCI) which utilizes benthic macroinvertebrates (BMIs), and Colilert testing for *Escherichia coli* (*E. coli*). Average water quality indices were calculated. Pigeon Hollow Run averaged 76.4 points (out of a possible 100), and Raven Rocks Run averaged 83.9 points. Both of these VSCI results represent excellent water quality. The water quality of Rocky Branch was categorized as good, averaging 69.2 points. *E. coli* samples were collected at two locations on each tributary a total of four times between June and December 2016. The *E. coli* count exceeded the Virginia State Water Control Board's most probable number (MPN) limit of 235 colonies/100 ml only once out of 24 times. Together, these data indicate that the Cool Spring campus area provides good to excellent water quality to the Shenandoah River via the site's three principal tributaries

A COMPARISON OF THREE POTABLE WATER SOURCES IN THE BLUE RIDGE MOUNTAINS OF VIRGINIA. Kaitlyn M. Dobyns, Katherine E. Plucinski & Kathy D. Gee, Dept. of Biological & Environmental Sciences, Longwood University, Farmville, VA 23909. Even in areas with ample annual rainfall, such as the Eastern U.S., access to freshwater is sometimes difficult. This is the case on Flat Top Mountain in Greene County, VA, where groundwater wells are unreliable and many properties do not have access to springs. In these situations, rainwater harvesting (RWH) can be an attractive option for communities. RWH systems collect rainwater and store it for later use. There is some concern, however, about their suitability for potable use and current regulations in Virginia make it difficult to use RWH as the sole source of potable water for a dwelling. This study analyzes the microbial quality of harvested rainwater, well water, and a freshwater spring. Our findings thus far support that harvested rainwater is of equal or superior quality to these alternative sources. (Supported by: Longwood University's Faculty Research Development Grant).

PERSISTENCE AND SUSTAINABILITY: TRACE MERCURY IN "UNCONTAMINATED" ROCKINGHAM CO. VA TERRESTRIAL ECOSYSTEMS REPRESENT A WIDESPREAD PATTERN OF OCCURRENCE. Gregory E. Mansour, Melissa E. Encinias, Matthew W. Riordan, Cody J. Cubbage & W. Dean Cocking, Dept. of Biol., James Madison Univ., Harrisonburg, VA 22807. Forest ecosystems in the western Shenandoah Valley of Virginia are not exposed to major sources of Hg contamination. Almost two decades ago Rockingham Co was assumed to be suitable as a low level control for comparison with sites exposed to major industrial contamination in Waynesboro VA. Mercury was detected at trace concentrations in soil, duff and airborne Hg samplers confirming that a control is not the same thing as a blank. The current study (2014-2017) examined four successional forest ecosystem sites with a focus on invertebrate organisms. Hot concentrated nitric and sulfuric acid digested sample replicates of composite passive air sampler residue, soil, and "duff" were analyzed for total Hg (THg) using Atomic Absorption Spectrophotometry (FIMS). Detectable trace amounts of Hg were present resulting in

suitable essentially replicate sites. This prompted the question of whether there is generally a detectable ambient THg presence in terrestrial habitats not exposed to known point sources. The literature shows that traces of THg are widely present in most control sites creating a useful baseline for monitoring future change.

REGIONAL AND GLOBAL SEA-SURFACE TEMPERATURES DURING THE LAST INTERGLACIATION. <u>Jeremy S. Hoffman</u>[1], Peter U. Clark[2], Andrew C. Parnell[3] and Feng He[2,4], [1]Science Museum of Virginia, Richmond, VA 23220, [2]College of Earth, Ocean, and Atmospheric Sciences, Oregon State University, Corvallis, OR 97330, [3]School of Mathematics and Statistics, University College Dublin, Dublin 4, Ireland and [4]Center for Climatic Research, Nelson Institute for Environmental Studies, University of Wisconsin-Madison, Madison, WI 53706. The last interglaciation (LIG, 129 to 116 thousand years ago) was the most recent time in Earth's history when global mean sea level was substantially higher than it is at present. However, reconstructions of LIG global temperature remain uncertain, with estimates ranging from no significant difference to nearly 2°C warmer than present-day temperatures. Here we use a network of sea-surface temperature (SST) records to reconstruct spatiotemporal variability in regional and global SSTs during the LIG. Our results indicate that peak LIG global mean annual SSTs were 0.5 ± 0.3°C warmer than the climatological mean from 1870 to 1889 and indistinguishable from the 1995 to 2014 mean. LIG warming in the extratropical latitudes occurred in response to boreal insolation and the bipolar seesaw, whereas tropical SSTs were slightly cooler than the 1870 to 1889 mean in response to reduced mean annual insolation. (Research was supported by an NSF Graduate Research Fellowship to J.S.H. under NSF grant 1314109-DGE and by NSF grants 1335197 and 1503032 to P.U.C. F.H. was supported by NSF grant AGS-1502990 and by the National Oceanic and Atmospheric Administration's Climate and Global Change Postdoctoral Fellowship program, administered by the University Corporation for Atmospheric Research.)

<u>Posters</u>

FORENSIC ANIMAL HAIR ANALYSIS: DEVELOPMENT OF AN ATLAS AND GUIDE. <u>Morgan A. Roth</u> and J. Thomas McClintock, Department of Biology and Chemistry, Liberty University, Lynchburg, VA 24502. Hair, collected as evidence from a crime scene, can be an important element in a criminal investigation since such evidence can demonstrate physical contact with a suspect(s), a victim(s), and a crime scene. When forensic scientists analyze hairs, the first step is determining if the hairs came from a human or an animal. Numerous resources are available for guidance on human hair analysis; however, minimum guides are available to identify and characterize animal hair. The objective of this research project was to compile a colored atlas and guide on the comparison of animal hair for use in criminal investigations. This atlas and guide will be comprised of colored photomicrographs of various animal hairs, as viewed through a forensic comparison microscope, assembled, labeled, and arranged into a guide for forensic use. Sketches of the photomicrographs will assist the viewer in hair structure and identification. Being able to correctly identify animal hairs could be very important from a forensic standpoint, as hair found at the scene of a crime could be crucial in apprehending the perpetrator. Animal hair identification is also important when viewed from a zoological standpoint, as hair can be an excellent aid in species identification.

LAND USE/LAND COVER (LU/LC) CHANGES IN VIRGINIA. <u>Devin Straub</u> & C.F. Labosier, Dept. of Biological & Environmental Sciences, Longwood University, Farmville, VA 23909. Land use/land cover (LULC) characteristics change over time through natural and human activities. Increased human activity has resulted in habitat destruction, species extinction, and landscape degradation by patch fragmentation, which influences biodiversity and climatic conditions at local and/or regional levels. A spatial and statistical analysis using data provided by U.S. Geological Survey was conducted for Prince Edward County, Virginia to determine LULC trends over a ten-year period: 2001, 2006, and 2011. After a reclassification of the Landsat-based, 30-meter resolution, raster data, a ten-year map series was created for the three periods in Prince Edward County. A statistical analysis showed that the study area showed a percent area decrease in six out of the fourteen LULC categories. Prince Edward experienced a 0.13% total increase in developed land, equating to 119.34 hectares of additional urbanization and a 4.15% decrease in total forest coverage, equating to a loss of 3,808.53 hectares of available forest habitat.

Medical Sciences

SYNTHESIS OF BIVALENT PROBES TO ELUCIDATE THE MECHANISM OF OPIOID ENHANCED NEUROAIDS. <u>Bethany A. Falls</u> & Yan Zhang, Department of Medicinal Chemistry, Virginia Commonwealth University, Richmond, VA 23284. AIDS (acquired immune deficiency syndrome) is a condition that occurs in the advanced stages of human immunodeficiency virus (HIV) infection. AIDS can lead to a wide variety of neurological disorders such as HIV- associated dementia and neuropathic pain. These associated neurological disorders are collectively referred to as neuroAIDS. The process of HIV infection is facilitated by co-receptors located on the surface of cells. One such co- receptor is the chemokine receptor CXCR4. This receptor is the entry receptor for the X4 virus, which emerges later in disease progression and has been associated with a more rapid progression to AIDS. Studies have shown that activation of the mu-opioid receptor (MOR) elevates the expression of CXCR4, thus enhancing viral invasion and a more rapid progression to neuroAIDS. This correlation suggests crosstalk between these two receptors; however, this mechanism of cross-talk is not very well understood. Therefore, to explore the possibility of one of these mechanisms, formation of MOR-CXCR4 heterodimers, we have successfully designed and synthesized a series of novel bivalent probes containing both the MOR antagonist naltrexone and the CXCR4 antagonist It1t. These ligands were designed to interact with both receptors synergistically. These ligands and their corresponding monovalent controls were synthesized in reasonable yields through synthetic routes established in our lab. The compounds were all fully characterized using ^{1}HNMR, ^{13}CNMR, MS, IR, and HPLC. Further biological characterization is underway to further apply them as chemical probes to study neuroAIDS.

A NOVEL AND PROMISING INHIBITOR OF THE NLRP3 INFLAMMASOME. <u>Jacob W. Fulp</u>[1], Chunqing Guo[2], Jeremy E. Chojnacki[1], Xia Li[2], Xiang-Yang Wang[2] & Shijun Zhang[1], [1]Dept. of Medicinal Chemistry, [2]Department of Human & Molecular Genetics, Virginia Commonwealth Univ., Richmond, VA 23284. Inflammasomes play a vital role in innate immunity by sensing microbial products and endogenous molecules associated with cellular damage and stress. The NOD-like receptor family pyrin domain containing 3 (NLRP3) inflammasome, is the most well studied inflammasome. Activation of the NLRP3 inflammasome triggers the production of the proinflammatory cytokines, interleukin (IL)-1β and IL-18. Production of (IL)-1β and IL-18

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promotes inflammation and pyroptosis. Notably, studies suggest that the dysregulation of the NLRP3 inflammasome plays important roles in the pathogenesis of a variety of diseases, including Alzheimer's disease, ischemia-reperfusion injury, obesity, atherosclerosis, type II diabetes and multiple sclerosis. Therefore, it is of great importance to develop novel NLRP3 inflammasome inhibitors, which will help further elucidate the roles of NLRP3 and (IL)-1β in the pathogenesis of inflammatory disorders, and which will ultimately facilitate the development of novel NLRP3 agents to combat these diseases. Recently, our lab has designed a novel small molecule NLRP3 inhibitor, JC171. *In vitro* characterization demonstrated that JC171 inhibits the production of (IL)-1β in J774.A1 macrophage cells upon challenge with LPS/ATP. More importantly, JC171 has been shown to be active in EAE mice, a mouse model of human multiple sclerosis in both preventive and therapeutic settings. (Funded by the NIA/NIH grant AG041161, Alzheimer's Drug Discovery Foundation, and the CHRB Award.)

MORPHOMETRY OF VISCERAL ADIPOSE TISSUE COMPOSITION AND ITS CORRELATIONS WITH MOLECULAR SIGNALING EVENTS AND PROGRESSION OF NON-ALCOHOLIC FATTY LIVER DISEASE (NAFLD). Joseph T. Bradley, Dept. of Biol., George Mason Univ., Fairfax, VA 22030. Nonalcoholic fatty liver disease (NAFLD) is the most common liver disease in the United States. The progression of NAFLD is associated with fat accumulation in the liver, increase in inflammation, increase in fibrosis, and resistance to insulin. Brown adipose tissue (BAT), found in the visceral adipose tissue (VAT), has properties that are inversely related to NAFLD. This experiment investigated whether the presence and activity of BAT is protective against NAFLD. To do this, protein products of BAT related genes from VAT biopsies were correlated with liver biopsies via computerized morphometry (CM). The amount of BAT in the VAT was measured by performing a genomic DNA to mitochondrial DNA ratio. Bead based immunoassay kits and Elisa kits were used to quantify the overall inflammatory state. In this study, TGF- β1 levels were found to be significantly negatively correlated with the degree of steatosis. (Supported by: Virginia Academy of Science.)

STUDY TO DETERMINE THE EFFECTS OF RESVERATROL ON THE BLOOD SERUM LEVELS OF ESTROGEN, ESTRONE, ESTRONE SULFATE AND ESTRADIOL IN FEMAIL PATIENTS WITH CYTOCHROME P450 GENETIC VARIATIONS. Yasamin Rahmani[1], Dr. Ancha Baranova[1] & Ruth Kriz APRN[2], [1]Department of Biology, George Mason University, Fairfax, VA 22030, [2] **Ruth Kriz, MSN, APRN**, Washington, DC 20008. Breast cancer is one of the four major cancers along with lung, colorectum and prostate. It is estimated that 1 in every 8 females will be affected by breast cancer in their lifetime. Certain genetic mutations, associated with estrogen metabolism, have been linked to the over production of metabolites that damage DNA leading to the formation of cancer cells. These metabolites have been linked to an imbalance of estrogens, specifically estrone (E_1), estrone sulfate (E_1S), and estradiol (E_2). With enzymatic activity of cytochrome P450 (CYP), CYP1A1 metabolizes estradiol into 2-hydroxyestradiol (2-OH-E_2), while CYP1B1 metabolizes both estradiol and estrone into 4-hydroxyestradiol (4-OH-E_2). Overproduction of 4-OH-E_2 has been linked to the formation of squamous cell carcinoma due to its binding to the DNA. This research is focused on investigating how resveratrol acts as a competitive inhibitor and slows down the enzymatic process of estrogen in Phase I of the liver detox pathway. Such properties of resveratrol have been observed *in vitro* but in this study, it will be taken a step further and observed *in vivo*. The study will observe the effect of resveratrol on the enzymatic activity of the cytochrome P450 enzyme variation of CYP1A1 and CYP1B1 in respect

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to the levels of estrogen, estrone, estrone sulfate and estradiol. It will also measure sex hormone binding globulin (SHBG) as an additional indicative of resveratrol's behavior *in vivo.*

AROMATIC ALDEHYDES AS ALLOSTERIC MODULATORS OF HEMOGLOBIN FOR THE TREATMENT OF SICKLE CELL DISEASE. Piyusha P. Pagare[1], Osheiza Abdulmalik[2], Martin K. Safo[1] & Yan Zhang[1], [1]Dept. of Med. Chem., Virginia Commonwealth Univ., Richmond, VA and [2]Children's Hospital of Philadelphia, PA. Intracellular polymerization of deoxygenated sickle hemoglobin (Hb) remains the principal cause of the pathophysiology associated with sickle cell disease (SCD). Allosteric effectors of Hb have been investigated as potential therapeutic agents for SCD. Our previous studies have shown that aromatic aldehydes form high-affinity Schiff-base adducts with Hb and inhibit red blood cell (RBC) sickling by increasing the oxygen affinity of Hb. However, challenges related to short half-life have severely hampered this line of investigation. To counter these challenges, we designed and synthesized a series of compounds based on our previously reported derivatives of vanillin followed by the in vitro investigations to ascertain sustained binding, modification of Hb and sickling inhibition. Our results showed maximal levels of Hb modification at 12 h, which were sustained for 24h suggesting that our modifications limit drug metabolism in RBCs. The compounds also showed inhibition of RBC sickling up to 100%. To establish the mode of interaction with Hb, we conducted x-ray crystallography studies, which showed that these compounds bind symmetrically at the α-cleft of Hb and show additional interactions with the F-helix which is known to be involved in polymer stabilization. Thus, our results establish these compounds as a novel, promising group of potent anti-sickling agents, demonstrate their proposed mechanism of action, and provide proof-of-concept justifications for our structure-based approach to developing potent therapeutics for SCD.

TARGETING NICOTINIC ACETYLCHOLINE RECEPTORS FOR THE PREVENTION AND REVERSAL OF CHEMOTHERAPY-INDUCED PERIPHERAL NEUROPATHY. S. L. Kyte[1], W. Toma[1], J. W. Bigbee[2], M. I. Damaj[1] & D.A. Gewirtz[1], [1]Dept. of Pharmacology and Toxicology and [2]Dept. of Anatomy and Neurobiology, Virginia Commonwealth Univ., Richmond VA 23298. Chemotherapy-induced peripheral neuropathy (CIPN), a consequence of nerve fiber dysfunction or degeneration, is characterized by sensory symptoms such as allodynia, resulting in a decreased quality of life. Paclitaxel, a taxane commonly used to treat breast, lung, and ovarian cancers, causes CIPN in 59-78% of cancer patients. Our data indicate that the nicotinic acetylcholine receptor (nAChR) agonist nicotine may be a potential preventative and therapeutic treatment for CIPN. Male C57BL/6J mice received paclitaxel every other day for a total of four injections (8 mg/kg, i.p.). The von Frey test revealed that nicotine can prevent (24 mg/kg/day, s.c.) and reverse (0.3-0.9 mg/kg, i.p.) paclitaxel-induced mechanical allodynia; the latter of which can be inhibited by the nAChR antagonist mecamylamine (2 mg/kg, s.c.). Chronic nicotine treatment can also prevent paclitaxel-induced intra-epidermal nerve fiber loss. In addition, nicotine (1 μM) fails to enhance viability, colony formation, and proliferation of A549 and H460 lung cancer cells. Also, nicotine (1 μM) does not interfere with paclitaxel (50-100 nM)-induced decreases in colony formation and proliferation, but modestly decreases the apoptotic A549 population. Finally, nicotine (24 mg/kg/day, s.c.) does not enhance tumor growth in Lewis lung carcinoma-bearing C57BL/6J mice. These data suggest that nAChRs may be promising drug targets for the prevention and treatment of CIPN. (Supported by: VCU Massey Cancer Center and National Institutes of Health.)

ALTERNATIVES TO POLYNUCLEOTIDE KINASE AND PHOSPHATASE (PNKP) ENZYME IN PROCESSING THE DNA DOUBLE-STRAND BREAKS. <u>Sri Lakshmi Chalasani</u>[1], Ajinkya Kawale[1], Mesfin Fanta[2], Michael Weinfeld[2] & Lawrence F Povirk[1], [1]Massey Cancer Center, VCU, Richmond VA 23298, [2] Cross Cancer Institute, Cross Cancer Institute, University of Alberta, Edmonton, Alberta, T6G1Z2. The enzyme polynucleotide kinase phosphatase (PNKP) plays a crucial role in a variety of DNA repair pathways by rendering the strand breaks ligatable. PNKP loss has been implicated in radiosensitivity, chemosensitivity and neurological disorders. PNKP has the ability to phosphorylate 5′-OH and dephosphorylate 3′-PO4 DNA ends by its kinase and phosphatase domains respectively. Phosphatase activity of PNKP is predominant than its kinase activity. Previous work in our lab has shown that cells with either PNKP knockdown or treated with A12B4C3 (PNKP phosphatase inhibitor) had little effect on 3′-phosphate kinetics. In the current study we employed PNKP knockout cells instead to analyze the 3′-phosphatase activity. In clonogenic survival assays, PNKP /- cells showed ~2.4-fold hypersensitivity to neocarzinostatin, which specifically induces 3′ phosphate- and 3′ phosphoglycolate-terminated DSBs. Furthermore, neocarzinostatin-induced DSBs, detected as 53BP1 foci, were more persistent in PNKP /- cells. However, in-vitro studies using different DSB substrates terminated with 3′-phosphate on one strand had their phosphate removed (70% in overhangs and 10% in recessed ends) despite the PNKP knockout status of the extracts. Further studies using these extracts will be performed to test for the presence of kinase activity in the absence of PNKP. Role of other weak 3'-DNA phosphatases in the context of NHEJ will also be evaluated.

ANAPLASMA PHAGOCYTOPHILUM INFECTION MODULATES EXPRESSION OF MEGAKARYOCYTES CELL CYCLE GENES THROUGH PHOSPHATIDYLINOSITOL-3-KINASE SIGNALING. <u>Supreet Khanal</u>[1], Hameeda Sultana[1, 2], John D Catravas[3, 4], Jason A. Carlyon[5] & Girish Neelakanta[1, 2], [1]Department of Biological Sciences, [2]Center for Molecular Medicine, College of Sciences, [3]Frank Reidy Research Center for Bioelectrics, [4]School of Medical Diagnostic and Translational Sciences, College of Health Sciences, Old Dominion University, Norfolk, VA, [5]Department of Microbiology and Immunology, Virginia Commonwealth University Medical Center, Richmond, VA 23298. *Anaplasma phagocytophilum*, the agent of Human Granulocytic Anaplasmosis infects neutrophils and other cells from hematopoietic origin. Using MEG-01 cell line, we show that expression of cell cycle genes in these cells are altered upon *A. phagocytophilum* infection. Expression of several cell cycle genes in MEG-01 cells was significantly up regulated at early and then down regulated at later stages of infection. Lactate dehydrogenase assay revealed reduced cellular cytotoxicity in MEG-01 cells upon *A. phagocytophilum* infection. The levels of both PI3CKA (p110 alpha-catalytic subunit) and PI3KR1 (p85-regulatory subunit) of Class I PI3 kinases and phosphorylated protein kinase B (Akt/PKB) and inhibitory kappa B (IκB) were elevated at both early and late stages of infection. Inhibition of PI3 kinases resulted in significant reduction in the expression of cell cycle genes, *A. phagocytophilum* burden and phosphorylated Akt levels in these cells. Collectively, these results suggest a role for PI3K-Akt-NF-κB signaling pathway in the modulation of megakaryocytes cell cycle upon *A. phagocytophilum* infection. (Supported by: Old Dominion University Startup funds to GN.)

DIET-INDUCED ALTERATION OF COX2 GENE EXPRESSION IN THE SPRAGUE DAWLEY RAT. <u>Jessica K. Morris</u> & Julia A. Halterman, MA in Biomedicine, Eastern Mennonite

Univ., Harrisonburg, VA 22801. Cyclooxygenase-2 (COX2), a product of the arachidonic acid metabolic pathway, plays a crucial role in the generation of the inflammatory response in mammals. Research has shown that increased COX2 gene expression, influenced by cytokines, growth factors, and other elements, such as diet, may contribute to proliferative diseases, such as cancer. The purpose of this study was to investigate the effects of a high-salt diet and a high-fructose diet on the expression of Cyclooxygenase-2 (COX2) in various tissues of Sprague Dawley rats. It was hypothesized that rats on a high-salt diet or a high-fructose diet would show an increased expression of the COX2 gene. Real-time PCR was used to amplify cDNA samples from 16 different body tissues to determine the level of COX2 expression in each tissue. The data was analyzed using a one-way ANOVA test. There was significant up-regulation of COX2 gene expression in adipose tissue and the kidney medulla for rats on a high-salt diet. On the other hand, there was significant down-regulation of COX2 gene expression in the aorta, skeletal muscle, stomach, intestine, and brain for rats on a high-salt diet and high-fructose diet. Based on these results, it can be concluded that high-salt and high-fructose diets may have an impact on COX2 gene expression in some tissues of the body. The effects of high-salt and high-fructose diets on COX2 gene expression should be investigated further to examine the chronic implications that may occur as a result of altered COX2 gene expression in various tissues of the body. (Funding source: The Thomas F. and Kate Miller Jeffress Memorial Trust, Bank of America, Trustee.)

ALTERNATIVE SPLICING OF NINEIN AS A FACTOR IN ANXIOLYTIC RESPONSES TO ETHANOL. Jessica L. Jurmain & Michael F. Miles, Dept. Pharm. & Tox., Va. Commonwealth Univ., Richmond, VA 23298. Anxiety has long been accepted as a factor in the initiation of alcohol abuse and occurrence of relapse associated with alcohol use disorders (AUD). Our laboratory recently identified a behavioral quantitative trait locus (QTL) in the nucleus accumbens influencing anxiolytic-like responses to acute doses of ethanol in the light-dark transition model of anxiety in BXD recombinant inbred mice (*Etanq1*). Mixed model analyses and fine mapping of the *Etanq1* support interval identified *Ninein* (*Nin*), which encodes a centrosomal protein that interacts with GSK3β, as a suggestive quantitative trait gene underlying anxiolytic-like responses to ethanol. qRT-PCR and western blots identified differences in expression of *Nin* and two provisional Ninein (NIN) protein isoforms between C57BL/6J (B6) and DBA/2J (D2) progenitor strains. Analyses of single nucleotide polymorphisms (SNPs) between B6 and D2 mice indicate that a strain specific difference in *Nin* sequence could underlie alternative splicing of *Nin* transcripts that encode proteins corresponding in molecular weight to the provisional protein isoforms identified in previous experiments. Recent literature suggests that differences in splicing of these transcripts results in functionally different NIN isotopes that specifically localize to the cytoplasm of mature neurons. We identified differences in expression of these transcripts, previously assumed to be targets of nonsense-mediated mRNA decay, between B6 and D2 mice using qRT-PCR. These results suggest structural and functional differences in NIN may modulate anxiolytic-like responses to ethanol.

Posters

THE ROLE OF NICOTINIC ACETYLCHOLINE RECEPTORS ON FREE-CHOICE ORAL CONSUMPTION OF NICOTINE. Deniz Bagdas[1,2] & M. Imad Damaj[1], [1]Department of Pharmacology and Toxicology, Virginia Commonwealth University, Richmond, VA and [2]Experimental Animals Breeding and Research Center, Uludag University, Bursa, Turkey.

Although the rate of smoking has decreased in the last decade, the smoke-free laws in the United States induced the alternative marketing of nicotine. There is an increasing interest to use oral nicotine delivery systems such as strips, orbs, sticks, and lozenges. Therefore, we aimed to determine the role of nAChRs on oral nicotine consumption in mice. Adult genetically modified mice, mice carrying deletions for the certain nicotinic acetylcholine receptor (nAChR) subunits, (n=8-10/per group/per sex) were given a choice of water or nicotine (a range of concentrations of 10-120 µg/ml) solution using two bottle free choice drinking assay. Our results showed that nicotine consumption varies in genetically modified mice due to subunit of nAChRs. In general, we observed that female mice significantly consumed nicotine more than male mice in wild-type (WT) littermates of knock-out (KO) mice. While the mice lack of β2 nAChRs showed significant decrease on nicotine intake at all concentrations, deletion of α5 nAChRs induced increase on nicotine consumption at high concentrations. The nicotine intake in male α6 KO mice was significantly higher than females. α6 KO female mice also consumed lesser than their WT controls. In summary, β2, α5 and α6 nAChRs may play a key role in nicotine intake and reward. In addition, the effects of nAChRs on nicotine intake and reward were sex-dependent. (This study was supported by Center for the Study of Tobacco Products, Virginia Commonwealth University.)

DEVELOPMENT OF NLRP3 INFLAMMASOME INHIBITOR FOR AD. Ashley G. Boice, Shijun Zhang, Jeremy Chojnacki, Liu He & Jacob Fulp, Virginia Commonwealth University, Richmond, VA.   Alzheimer's disease (AD) is a progressive neurodegenerative disease characterized by memory loss and dementia. AD affects millions of Americans and is detrimental to the individual and family affected as well as the rising health care cost as the number of elderly people in our population increases. One therapeutic strategy for this disease is targeting the NLRP3 inflammasome to decrease the neuroinflammation of this disease that contributes to cell death and the devastating cognitive effects. NLRP3 inflammasome is a multiprotein complex that consists of proteins caspase-1, ASC, and NLRP3. NLRP3 senses damage or pathogen associated molecular patterns, leading to the activation of caspase-1 and subsequent cleavage of pro-IL-1-beta and pro-IL-18 into their active form IL-1-beta and IL-18. A recent study found that the anti-diabetic drug, glyburide, inhibits NLRP3, but the dose necessary for inhibition of NLRP3 inflammasome would cause lethal hypoglycemia. A glyburide analog with the removal of the cyclohexylurea moiety necessary for anti-diabetic activity, GA3, was found to inhibit caspase-1 activity as well as mitigate damaging inflammatory effects in murine models. Further optimization of GA3 has led to more potent leads such as HL-12. In order to uncover the binding site of GA3 and analogs, Microscale Thermophoresis (MST) assays were conducted on GA3, HL-12, HL-16 and known NLRP3 inflammasome inhibitor MCC950 for the evaluation of binding affinity to proteins of the NLRP3 inflammasome complex: NLRP3 and ASC. (Supported by: Alzheimer's Drug Discovery Foundation.)

NOVEL INHIBITOR OF THE NLRP3 INFLAMMASOME INHIBITS THE PRODUCTION OF (IL)-1β AND DISPLAYS PROMISING THERAPEUTIC PROPERTIES IN MULTIPLE SCLEROSIS MOUSE MODEL. Jacob W. Fulp[1], Chunqing Guo[2], Jeremy E. Chojnacki[1], Xia Li[2], Xiang-Yang Wang[2] & Shijun Zhang[1], [1]Dept. of Medicinal Chemistry, [2]Department of Human & Molecular Genetics, Virginia Commonwealth Univ., Richmond, VA 23284. We have developed a novel and potent inhibitor of the NOD-like receptor family pyrin domain containing 3 (NLRP3) inflammasome as a potential therapeutic for the treatment of multiple sclerosis (MS). This compound, known as JC-171, was designed using the scaffold of glyburide. Our studies indicated

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that JC-171 dose dependently inhibited the secretion of interleukin-1β (IL-1β) from J774A.1 macrophage cells under LPS/ATP challenge. The IC50 value was determined to be 8.45 ± 1.56 μM. Additionally, the inhibition of IL-1β secretion was also confirmed using mouse bone marrow derived macrophage cells and mice that were subjected to LPS-challenge. More importantly, the administration of JC-171 delayed the onset, severity, and progression of experimental autoimmune encephalomyelitis (EAE), a mouse model of MS. This therapeutic effect was associated with a reduction in the production of IL-1β. Together, these results imply that JC-171 is a selective NLRP3 inflammasome inhibitor with biological activity in both an *in vitro* and *in vivo* settings. These findings strongly encourage further optimization of this molecule as a potential therapeutic agent for MS. (This project was funded by the NIA/NIH grant AG041161, Alzheimer's Drug Discovery Foundation, and the CHRB Award, Commonwealth of Virginia.)

DOES METHYLENE BLUE TREATMENT IMPACT OLFACTORY AND COGNITIVE DEFICITS IN A TRANSGENIC MOUSE MODEL OF ALZHEIMER DISEASE? Harold J. Grau, Benjamin C. Genovese, Nazifa Khan, Emma Croushore, Katie Whitcomb, Rachel A. Schendzielos, Darlene A. Mitrano & Lisa S. Webb, Dept. of Mol. Biol. & Chem., & Program in Neurosci., Christopher Newport Univ., Newport News, VA 23606. Alzheimer Disease (AD) is a degenerative form of dementia that is associated with the accumulation of neurofibrillary tangles (tau protein) and amyloid-beta (AB) plaque formation. Methylene blue (MB) has been FDA approved for reducing protein aggregations in AD and other diseases. In this study, we used a triple transgenic mouse model of AD (3xTg-AD) to assess any effects of MB on the formation of these protein tangles and deposits (reported elsewhere) and on spatial learning and memory tasks (reported here), by comparing mice given weekly intraperitoneal injections of MB with those given saline injections (controls). We used the Morris Water Maze (MWM) to test for spatial learning and memory on mice at 3, 4.5, 6, 7.5, 9, 12, 15, and 18 months of age; mice were given 5 days of training (with platform), and then tested (no platform) on day 6. While MB treatment had some positive effects, the only significant (P<0.05) effect of MB on training was at 4.5 months, where the MB group reached criterion (less than 25 sec in at least 4/5 trials) in fewer average days than the saline controls. Similarly, MB did not seem to significantly enhance 24-hour spatial memory, except in the 3-month cohort. Small sample sizes, a consequence of attrition that was likely exacerbated by the weekly injection, prevented many of the differences found from being statistically significant. Olfactory deficits were tested with a peanut butter choice test, but produced inconclusive results.

NFAT5 IS DIFFERENTIALLY EXPRESSED IN SPRAGUE-DAWLEY RAT TISSUES IN RESPONSE TO HIGH SALT AND HIGH FRUCTOSE DIETS. Braden Herman, Samantha Kauffman, Marchelle Smucker, & Julia Halterman, Dept. of Biol., Eastern Mennonite Univ., Harrisonburg VA 22802. In the current diet of an average American, there is an increasing amount of both salt and high fructose corn syrup. Individuals with a higher sensitivity to salt or fructose can develop hypertension and metabolic syndrome. In this study, we aimed to determine how consumption of a high salt or a high fructose diet alters tissue-specific expression of the NFAT5 gene. Fifty rats were put on a control, 4% NaCl, 8% NaCl, or 64% fructose diet over the course of 8 weeks. Sixteen tissue samples were then harvested from the rats. RNA was isolated and purified from the samples, and reverse transcription was used to convert purified RNA into cDNA. RT-PCR was used to measure expression of the NFAT5 gene. An increase in NFAT5 expression was found in the kidney medulla on the 4% NaCl diet, in the kidney cortex on the 64% fructose diet,

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in the adipose tissue with the 4% and 8% NaCl diets, in the liver with the 8% NaCl diet, and in the brain with the 4% NaCl diet. A decrease in NFAT5 expression was found in the bladder with the 8% NaCl and 64% fructose diets, and in the spleen with the 64% fructose diet. There was no significant difference in NFAT5 expression in the pancreas, lungs, skin, small intestine, blood, aorta, heart, stomach, or skeletal muscle. Based on these results we can conclude that NFAT5 is differentially expressed in different tissues in response to various diets. (Supported by: The Thomas F. and Kate Miller Jeffress Memorial Trust, Bank of American Trustee.)

KINETICS AND INHIBITOR STUDIES OF THE L205R-PKACα MUTANT. N. Luzi, D. Peterson, & K. C. Ellis, Dept. of Med. Chem., Virginia Commonwealth Univ., Richmond VA 23298. Cyclic-AMP dependent protein kinase (PKA) is involved in many biological processes and is made up of two regulatory and two catalytic subunits. When the catalytic subunits are released they participate in various downstream effects, including the production of cortisol. When PKACα undergoes a single amino acid change in the substrate-binding site, L205R-PKACα, the protein becomes constitutively active leading to increased cortisol production in a subset of ACTH-Independent Cushing's Syndrome patients. Our goal is to develop selective inhibitors of the L205R-mutant. However, in order to do so we need to study the kinetics and inhibition from known PKA inhibitors of both wt- and L205R-PKACα prior to testing novel compounds. The kinetics of the substrate-binding site for both wt- and L205R-PKACα were determined using a novel, fluorophore-labeled substrate peptide in a endpoint assay followed by detection of substrate and product by HPLC. The K_M[peptide] for L205R-PKACα was 60.68 ± 7.082 μM, which was 6-fold higher compared to wt-PKACα where K_M[peptide] = 9.74 ± 0.88 μM. Work to determine the K_M for the ATP-binding site for both proteins will also be presented. In our inhibition studies using the novel, fluorophore-labeled substrate peptide, the IC_{50} of H89 was determined to be 43.05 nM and the IC_{50} of PKI(5-24) was determined to be 8.45 nM. We will also present data for known PKA inhibitors against the mutant and the development of novel inhibitors selective against the mutant protein.

CELLULAR DEACETYLASE SirT1 REGULATES THE HPV16 REPLICATION COMPLEX. Valerie Marley[1], Dipon Das[1], Xu Wang[1] and Iain M Morgan[1,2]. [1]VCU Philips Institute for Oral Health Research, School of Dentistry, [2]VCU Massey Cancer Center. Human papillomaviruses (HPV) are causative agents in almost all cervical carcinomas and in head and neck cancer (HNC), the cases of which are increasing rapidly worldwide; HPV16 is causative in 50% of cervical and 90% of HPV+ HNCs. There is no therapeutic vaccine for managing HPV infections, only prophylactic vaccines, therefore, novel therapeutic targets are required to combat HPV disease. One such target is viral DNA replication which is carried out by the viral proteins E1 and E2 in association with cellular proteins. Here we describe the role of the cellular deacetylase SirT1 in the regulation of HPV16 E1 and E2 acetylation, protein levels, and replication function. SIRT1 is a cellular complex with E1 and E2 and is also recruited to the viral origin of replication in an E1-E2 dependent manner. In the HPV-negative cervical cancer cell line C33a, the SirT1 gene was removed by CRISPR. In the CRISPR knock-out cells the levels of E1 and E2 protein are elevated and the DNA replication function of these proteins is enhanced compared with wild-type C33a cells. Restoration of SirT1 to the CRISPR cells restores E1-E2 replication to levels observed in wild-type C33a cells. The conclusion from this work is that the enzyme SirT1 plays a crucial role in controlling HPV16 E1-E2 mediated DNA replication via controlling the acetylation and protein levels of these viral proteins. There are inhibitors and agonists of SirT1 that are currently in a

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clinical trial targeting a variety of cancers. We propose that such drugs may also be used to target and disrupt HPV16 infection and therefore assist in the battle against HPV-related cancers.

KAPPA OPIOID RECEPTORS MAY MEDIATE PACLITAXEL-INDUCED CHANGES IN AFFECT-LIKE BEHAVIOR IN MALE MICE. Julie A. Meade, Wisam B. Toma, Yasmin Alkhlaif, Dana E. Selley, & M. Imad Damaj, Dept. of Pharmacology & Toxicology, Virginia Commonwealth University, Richmond VA 23298. Longitudinal studies of cancer survivors show long-term changes in mood, such as dysphoria and emotional deficit, including anhedonia. In order to study the underlying mechanisms of long-term changes in mood induced by cancer chemotherapy, cancer-free male C57BL/6J mice were treated with one cycle of four injections of vehicle or the chemotherapeutic paclitaxel (32mg/kg cumulative) and periodically assessed for anhedonia-like behavior. Paclitaxel caused significant, time-dependent deficits in sucrose preference and morphine conditioned place preference. The selective kappa opioid receptor (KOR) antagonist norbinaltorphimine reversed paclitaxel-induced sucrose preference deficit, suggesting an important role for the KOR neuronal system in paclitaxel-induced anhedonia. Because KOR signaling in the nucleus accumbens (NAc) can cause anhedonia, we used the $[^{35}S]GTP\gamma S$ assay to determine if paclitaxel modulated KOR activity in the NAc. Surprisingly, a history of paclitaxel had a trend of reducing U50,488H-stimulated KOR activity. Our behavioral and *in vitro* data suggest that paclitaxel-induced changes in affect-like behavior may be due to dysregulation of KOR signaling in the limbic system (Supported by: NIH grant R01-CA206028, the VCU Massey Cancer Center, and NIH grant P30 CA016059.)

STRUCTURE-ACTIVITY RELATIONSHIPS IN NON-HEPARIN ANTICOAGULANTS. Shravan Morla, Elsamani Abdelfadeil, Daniel K. Afosah & Umesh R. Desai, Department of Medicinal Chemistry and Institute for Structural Biology, Drug Discovery and Development, Virginia Commonwealth University, Richmond, VA 23219. We recently discovered that a novel non-saccharide glycosaminoglycan mimetic, named G2.2, demonstrates highly selective cancer stem-like cells (CSCs) inhibition activity. G2.2 inhibited the growth of CSCs from multiple cancer cell lines. To assess its in vivo anti-cancer potential, we requested a contract research organization (CRO) to synthesize 20 g of material, labelled as G2.2C. Evaluation of G2.2C in HT-29 xenograft mouse model showed a significant reduction in tumor volume and CSC markers, and unexpected bleeding consequences in some animals. Similar studies with G2.2 synthesized in our laboratory had shown no bleeding effects. To investigate the difference between the two G2.2 samples (G2.2L (lab) and G2.2C (CRO)) we performed UPLC-MS characterization and found that G2.2C sample was a 85:15 mixture of two compounds. Elemental, NMR and MS data showed that G2.2L was fully sulfated flavonoid derivative, as expected, but G2.2C contained one less sulfate group. We tested both agents for their inhibition of various coagulation factors and found that G2.2C inhibited FXIa almost 2-fold better than G2.2L. Further, activated partial thromboplastin time assay indicated that G2.2L approximately 3-4-fold less anticoagulant than G2.2C. This implies that loss of just one sulfate group could induce significant side effects. Such structure–activity relationship is important to understand if the in vivo metabolism of the agents leads to accumulation of de-sulfated products.

THE EFFECT OF DIET ON ALDOSE REDUCTASE AND NFAT5 EXPRESSION IN RAT TISSUES. Kaylee M. Ferguson, Braden Herman, & Julia A. Halterman, Department of Biology, Eastern Mennonite University, Harrisonburg VA 22802. This research investigates the effects of

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a 4% NaCl, 8% NaCl, 64% fructose, and control diet on the expression of Aldose Reductase (AR) in rat tissues, as well as the relationship of AR and Nuclear Factor of Activated T-Cells 5 (NFAT5). In human health, low salt diets lower the risk of cardiovascular disorders and low fructose diets lower the risk of metabolic disorders. In this study, the effects of salt/fructose diets on AR and NFAT5 expression were examined in 16 tissues of Sprague Dawley rats. This was done by purifying RNA, creating cDNA, and quantifying AR expression by real-time PCR. Adipose tissue, kidney medulla, brain, and liver tissues showed "salt sensitivity." Kidney medulla and brain showed increased expression of AR and NFAT5 following consumption of 4% NaCl diet. Liver showed increased AR and NFAT5 expression in the 8% NaCl diet. Adipose tissue showed AR upregulation in an 8% NaCl diet, and upregulated NFAT5 expression following 4% and 8% NaCl diets. This suggests that NFAT5 and AR are regulated in the same way in tissues that process osmolytes directly, such as the kidney medulla and liver. Further inquiry is needed to find the mechanisms of AR and NFAT5 regulation in the brain and adipose tissue. (Funding source: The Thomas F. and Kate Miller Jeffress Memorial Trust, Bank of America, Trustee.)

REVERSIBILITY OF THERAPY-INDUCED SENESCENCE IN NON-SMALL CELL LUNG CANCER AS A MODEL OF CANCER RECURRENCE. T. Saleh[1], E.K. Cudjoe[4], S.L. Kyte[1], S. C. Henderson[2], L.W. Elmore[3] and D.A. Gewirtz[1], [1]Dept. of Pharmacology and Toxicology; [2]Dept. of Anatomy and Neurobiology; [3]Dept. of Pathology; [4]Department of Pharmacotherapy and Outcomes Science, VCU, Richmond, 23298. Autophagy and senescence are both well-established responses to chemotherapy and radiation that often occur in parallel, contributing to growth arrest in tumor cells. However, it has not been established whether this growth arrest is reversible. This question was addressed using non-small cell lung cancer models exposed to the cancer chemotherapeutic drug, etoposide. Senescent cells that were sorted, identified by β-galactosidase staining and alterations in morphology, isolated by flow cytometric cell sorting based on C12FDG staining, and real-time live microscopy were found to be capable of recovering proliferative capacity. Autophagy, monitored by vacuole formation, p62 degradation, and LC3BII generation did not interfere with either the senescence arrest or proliferative recovery and was non-cytoprotective in function. These observations argue against the premise that (chemotherapy-induced) senescence is strictly irreversible and indicate that therapy-induced senescence may ultimately be a transient process in that at least a subpopulation of tumor cells can and will remain metabolically active and recover proliferative capacity independently of autophagic turnover. We therefore propose that dormant tumor cells may be capable of prolonged survival in a state of autophagy/senescence and that disease recurrence may reflect escape from this senescence-arrested state. (Supported by Massey Cancer Center and Office of the Assistant Secretary of Defense.)

COMPUTATIONAL IDENTIFICATION OF POTENTIAL mRNA BIOMARKERS INDICATIVE OF DOWNSTREAM HCC DEVELOPMENT IN NAFLD PATIENTS. Simran J. Singh, Sch. of Sys. Biol., GMU, Fairfax VA, 22030. It is widely accepted that cancer develops as a result of a combination of various mutations that allow a cell not only to replicate without regulation, but also migrate from its typical cellular environment in order to metastasize in other regions of the body. This study hopes to design an approach that can be used to identify and validate the roles of certain mutations that result in differential changes in gene expression between the liver cells of patients suffering from Non-Alcoholic Fatty Liver Disease (NAFLD) and those with Hepatocellular Carcinoma (HCC.) Utilizing this data, it may be possible to identify certain

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changes in gene expression that may be indicative of higher-risk development of HCC in the future. 150 total gene expression data sets obtained using the Affymetrix GeneChip Human Genome U133 Plus 2.0 array have been downloaded from the EMBL-EBI ArrayExpress database, and this raw data is currently being processed utilizing the *limma* package for Bioconductor in R. Following data processing, pathways that were overexpressed in high numbers of the HCC patients but in relatively low numbers for the NAFLD patients will be selected for in-vitro modeling. Via the use of CRISPR/Cas9 to implement mutations that would model these gene expression changes in healthy primary human hepatocytes, differences in cell morphology, cellular division and cellular proliferation (utilizing a transwell migration assay) will be recorded. The observation of significant changes in any of these parameters would indicate a shift towards the phenotypes typically associated with metastatic cancer, and thus the potential for a higher risk of HCC development.

ASSOCIATION OF SERUM CYTOKINES WITH LIVER FIBROSIS AS MEASURED BY COMPUTERIZED MORPHOMETRY ACROSS GRADES OF NAFLD. <u>Sasha Stoddard</u>[1,2], Lakshmi Alaparthi[3], Zachary D. Goodman[3], Aybike Birerdinc[2,3] & Ancha Baranova[1,2,3], [1]Sch. of Sys. Biol., GMU, Fairfax, VA, [2]CSCMD, GMU, Falls Church, VA, [3]Inova Health Sys., Falls Church, VA. Global prevalence of nonalcoholic fatty liver disease (NAFLD) is currently at 25%, and is expected to increase as a major public health concern due to the current worldwide obesity epidemic. The spectrum of NAFLD includes nonalcoholic steatohepatitis (NASH) and steatohepatitic hepatocellular carcinoma (SH-HCC). Not all patients with NAFLD progress to NASH, and the diagnosis and grading of NAFLD/NASH is dependent on the "gold standard" of invasive liver biopsy. Molecular mechanisms behind the development and progression of this disease are poorly understood, and this study attempts to correlate serum biomarkers with quantified fibrotic liver changes in NAFLD patients compared to diagnostic categories. Liver biopsies from obese non-NAFLD, NAFLD, and NASH patients were analyzed via computerized morphometry to quantify levels of steatosis and fibrosis. Results of immunoassays on serum samples indicate a negative correlation of IL-7 ($p<0.05$), G-CSF ($p<0.05$), and GM-CSF ($p<0.05$) with percent collagen of liver biopsies, but no significance when comparing these protein levels between diagnostic categories. Correlation of serum biomarker levels with amount of fibrosis as determined by computerized morphometry has the potential to be a more sensitive tool in the assessment of diagnostic and/or prognostic biomarkers in NAFLD.

INVESTIGATION THE ROLE OF α7 NICOTINIC ACETYLCHOLINE RECEPTORS (nAChRs) AS A POTENTIAL TARGET FOR THE TREATMENT OF CHEMOTHERAPY INDUCED PERIPHERAL NEUROPATHY (CIPN). <u>Wisam B. Toma</u>[1], S. Lauren Kyte[1], Julie A. Meade[1], Deniz Bagdas[1], Ganesh A. Thakur[3], John W. Bigbee[2], David A. Gewirtz[1] & M. Imad Damaj[1], [1] Dept. of Pharmacology and Toxicology and [2]Dept. of Anatomy and Neurobiology, Virginia Commonwealth University, Richmond, VA 23298, [3]Dept. of Pharmaceutical Sciences, Northeastern University, Boston, MA 02115. Chemotherapy-Induced Peripheral Neuropathy (CIPN) is a major dose-limiting side effect of chemotherapeutics, such as paclitaxel. CIPN symptoms include numbness, allodynia, and hyperalgesia. The objective of this study was to characterize the role of α7 nicotinic acetylcholine receptors (nAChRs), which are distributed in the pain transmission pathway, in the pathology of paclitaxel-induced CIPN in a mouse model. We investigated whether R-47, a selective silent agonist of α7 nAChRs, would reverse and prevent CIPN. α7 WT and KO mice were treated with paclitaxel (1 mg/kg, i.p.). R-47 (1, 5, 10 mg/kg, p.o.) was given to mice treated with paclitaxel (8 mg/kg, i.p.) to reverse CIPN; R-47 (10 mg/kg,

p.o.) was repeatedly given to prevent CIPN. Our results show that α7 KO mice develop a significantly lower mechanical threshold compared to WT mice. Additionally, R-47 both prevents and reverses paclitaxel-induced mechanical allodynia. Our data suggests that α7 nAChRs could be a potential target to prevent and treat CIPN. (This work was supported by NIH grant R01-CA206028 to MID and DAG, and partly supported by pilot funding from the VCU Massey Cancer Center.)

Natural History and Biodiversity

PHYLOGENETIC RELATIONSHIPS OF *NOCOMIS* AND *EXOGLOSSUM* SPECIES BASED ON MORPHOLOGY, BEHAVIOR, AND GENETICS. Eugene G. Maurakis[1,2] & Thomas M. Orrell[3]. [1]Biology Dept., University of Richmond, VA 23173, [2]Science Museum of Virginia, 2500 W. Broad St., Richmond, VA 23220, & [3]Smithsonian Institution, National Museum of Natural History, PO Box 37012, Washington, DC 20013. Objectives were to determine phylogenetic relationships among pebble nest-building species of *Nocomis* and *Exoglossum* using 16 behavioral, 26 morphological traits, and DNA sequence data of the nuclear recombination-activating gene (*RAG1*) and the mitochondrial cytochrome *b* gene where *Campostoma anomalum* was used as the out-group. Analysis of combined character datasets indicate the genus *Nocomis* is monophyletic, where the *N. biguttatus* species-group is the sister group to the clade comprised of the *N. leptocephalus* and *N. micropogon* species-groups, in which all species level relationships are resolved. *Exoglossum laurae* and *Exoglossum maxillingua* form a monophyletic group, the sister-group to the *Rhinichthys* and *Oregonichthys* clades. When analyzed alone, *RAG1* data contributed little to unraveling relationships among all 16 species examined except for four species of *Nocomis* (*N. leptocephalus*, *N. raneyi*, *N. micropogon*, and *N. platyrhynchus*). In contrast, sequence data of cyt *b* singularly resulted in defining each of *Nocomis* and *Exoglossum* as monophyletic groups. Likewise, behavioral and morphological datasets resulted in defining each of *Nocomis* and *Exoglossum* as monophyletic groups. Our future studies are examining phylogenetic relationships of populations of *N. leptocephalus*, currently designated as subspecies (i.e., *leptocephalus*, *interocularis*, and *bellicus*).

POPULATION ECOLOGY OF THE RED-BACKED SALAMANDER AT THE SOUTHERN RANGE EDGE. Sarah Timko[1], Christian Law[1], Alexander Novarro[2], Lily Thompson[1] & Kristine Grayson[1], [1]Department of Biology,
University of Richmond, Richmond, VA 23173 and [2]Department of Biology, University of Maryland, College Park, MD 20742. Eastern red backed salamanders (*Plethodon cinereus*) are widely distributed throughout northeastern North America, with the southern portion of the range extending to Virginia and North Carolina. This study characterizes the activity, spatial dynamics, and critical thermal (CT) limits of a population of *P. cinereus* in an urban park in Richmond, Virginia. We established six artificial cover board plots and used count and mark-recapture survey methods to regularly monitor salamander activity over a year. Local abundance of *P. cinereus* in the James River Park System was surprisingly high for an urban, coastal plain population, with mean local densities of 1.96 – 2.03 adults per m^2. Spatial capture-recapture space-use estimates were 3.13–5.08 m^2 for females and 5.21–6.83 m^2 for males. The activity period of *P. cinereus* in this location extended from October to May, with surface activity only decreasing from June to September. CT limits were defined as the temperature at which an individual lost the ability to right itself. Minimum and maximum CT limits were quantified for the Richmond population and

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compared to three other populations across the latitudinal range extent. There were no differences in CT maximum across the populations, but CT minimum decreased as the mean number of days below 5°C decreased. Together, these results indicate that the thermal environment places important constraints on salamander activity.

LONG-TERM SURVIVORSHIP IN TWO EASTERN RED SPOTTED NEWT POPULATIONS WITH PARTIAL MIGRATION. Kayla Sherman[1], Andrew Levorse[1], Kristine Grayson[1] &; Larissa Bailey[2], [1]Department of Biology, University of Richmond, Richmond, VA 23173 and [2]Department of Fish, Wildlife, and Conservation Biology, Colorado State University, Fort Collins, CO 80523. Pond-breeding amphibians migrate to wetlands for breeding and then typically return to terrestrial habitats. When permanent aquatic habitats are available, species may overwinter in the ponds. This strategy, partial migration, is found in a range of species where populations are composed of residents and migrants. The Eastern red-spotted newt (*Notophthalmus viridescens*) is known for its life-cycle plasticity and it exhibits partial migration. Here we returned to two newt populations at Mountain Lake Biological Station ten years after individuals were first marked. We determined if known individuals remained and provided long-term estimates of survival trade-offs between migration and residency. We used multi-state mark recapture models to determine survival probabilities. We captured 65 known individuals and model rankings support the hypothesis that residents have higher long-term survival. New estimates were similar to projections from previous data, indicating conditional life history differences remained between migrants and residents. Additionally, we found that newts can persist for at least 10 years in the adult stage, resulting in impressive longevity given that the juvenile red eft stage can range from 2 – 8 years. Overall, this study provides evidence of long-term survival and migratory trade-offs in a common amphibian, where its site fidelity and tractability provides one of the longest running examinations of the mechanisms of partial migration.

FOSSIL REPTILES FROM THE MIOCENE OF VIRGINIA AND THE ECOLOGY OF A 14-MILLION-YEAR-OLD MARINE ECOSYSTEM. Alexander K. Hastings, Virginia Museum of Natural History, Martinsville, Virginia 24112. Roughly 100 km from the preset-day shoreline, the Carmel Church Quarry (CCQ) fossil locality in central Virginia preserves a section of marine deposits with a diverse fauna, primarily of sharks and cetaceans. Little has been published from this site regarding reptiles. Two units from this exposure have yielded herpetofauna: the Eocene Nanjemoy Formation and the Miocene Calvert Formation. The older Nanjemoy has yielded 4 isolated vertebrae of *Palaeophis*, an extinct marine snake known from other shallow near-coastal environments. Fossils recovered from the younger Calvert include 32 crocodylian teeth, 3 vertebrae and 6 un-keeled osteoderms that are consistent with *Thecachampsa*, known from other near-shore environments. Sea turtles are the most common fossil reptiles in the Calvert, with taxa that are also consistent with Miocene faunas. These include mostly portions of carapace (n = 119), but also at least 7 appendicular elements from *Syllomus, Procolpochelys and Psephophorus*. Of these, all but *Psephophorus* were likely inhabitants of near-coastal shallow environments. Pelagic or deep water taxa are rare (as well as terrestrial taxa), but do indicate a mix of environments represented at CCQ.

Posters

SMALL MAMMALS FROM THREE MOUNTAIN RANGES IN NUCLEAR CENTRAL AMERICA. <u>Ralph P. Eckerlin</u>[1], John O. Matson[2], Sergio Pérez[3] & Nicté Ordóñez-Garza[4], [1]Math, Science & Engineering Division, Northern Virginia Community College, Annandale, VA 22003, [2]Department of Biological Sciences, San Jose State University, San Jose, CA 95192, [3]Museo Historia Natural de la Universidad de San Carlos, de Guatemala, [4]Department of Biological Sciences, Texas Tech University, Lubbock, TX 79409. Small mammal ecology and natural history are poorly known in Nuclear Central America. In an effort to gain information on small mammals (insectivores, marsupials and rodents), we sampled 3 cloud forest habitats in mountain ranges in Honduras (Cerro Celaque and Cerro Agalta) and Guatemala (Sierra de las Minas). Small mammals were collected by removal trapping. Trapping effort varied among sites with trap-nights totaling 18117. A total of 789 specimens of 21 species of small mammals was recorded from the 7 trapping sites. Habitat at each trapping site is described. Species diversity, relative abundance, sex ratios, reproductive activity, and other natural history information are reported. In general, cloud forests in the 3 mountain ranges contained different communities of small mammals. Three species (*Heteromys desmarestianus*, *Peromyscus oaxacensis* and *Scotinomys tequina*) occurred in the cloud forests of all three mountain ranges. New species of *Peromyscus*, *Sorex*, and *Cryptotis* have been described.

EVALUATION OF AGONISTIC INTERACTIONS BETWEEN NATIVE AND INVASIVE CRAYFISH USING NOVEL REAL-TIME VIDEO TRACKING SYSTEM. <u>E. Lewis</u>, <u>J. Cruz</u>, <u>C. Milliron</u>, B. Jackson, & S. Henkanaththegedara, Department of Biological & Environmental Sciences, Longwood University, Farmville, VA 23909. The Eastern United States is the global hotspot for crayfish diversity with more than 440 species. Many of these native crayfish are threatened with extinction due to invasive crayfish species. In a sympatric environment, invasive species of crayfish have the ability to take over and replace native species rapidly through predation and competition for food and shelter. We studied the potential impacts of invasive red-swamp crayfish (*Procambarus clarkii*) on native Piedmont crayfish (*Cambarus* sp. C) using a novel real-time video tracking system based on agonistic behavior. The results showed that invasive crayfish have significantly higher aggression levels than the native crayfish species (W = 37, p = 0.0449). The maximum backward velocity of native crayfish was significantly higher than that of invasive species, showing signs of submissive behavior (W = 32, p = 0.0205). Use of the novel real-time tracking system provides more flexibility to measure traits associated with crayfish agonistic behavior. These findings may help managers to find novel management options to address issues related to invasive crayfish impacts.

LAND MAMMALS OF THE VIRGINIA BARRIER ISLANDS. Raymond D. Dueser[1], John H. Porter[1], & <u>Nancy D. Moncrief</u>[2], [1]Department of Environmental Sciences, University of Virginia, Charlottesville, VA 22904, and [2]Virginia Museum of Natural History, Martinsville, VA 24112. The Virginia barrier islands are a dynamic natural laboratory for the study of ecological and evolutionary patterns and processes. We have studied the ecology, distribution, and genetics of non-volant mammals on these islands since the mid-1970s. We have trapped, tracked, and observed mammals on 30 barrier and marsh islands and at more than 25 locations on the adjacent mainland in Accomack and Northampton counties, Virginia. We also have assembled published and unpublished reports from other researchers who have studied mammals on the islands and the

Delmarva Peninsula, which is made up of the state of Delaware and parts of Maryland and Virginia. In this report, we identify 34 species of land mammals that are native to this region. Of those, 20 species have been observed on at least one island. In addition, we identify four non-domesticated, exotic, species that have been observed on at least one island.

A SURVEY OF FRESHWATER FISHES OF THE UPPER APPOMATTOX RIVER BASIN IN CENTRAL VIRGINIA. R. Osborne, M. Conner, D. Conner, C. Perry & S. Henkanaththegedara, Department of Biological & Environmental Sciences, Longwood University, Farmville, VA 23909. Biodiversity surveys are crucial to provide law makers and conservation practitioners with the necessary data to understand the complex relationship between humans and biodiversity. The Appomattox River is the largest tributary of the James River and is poorly surveyed for freshwater fish diversity. During the summer of 2016, we sampled 31 sites within the upper Appomattox basin using dip nets and seines. Over 700 specimens were collected and identified to 9 different families and 36 species. Of these species we were able to identify three new species to the Appomattox River: White Shiner (*Luxilus albeolus*), Spottail Shiner (*Notropis hudsonius*) and Rock Bass (*Ambloplites rupestris*). We found cyprinids (86%), percids (76%) and centrarchids (62%) in more than half of the locations sampled. In comparison with low-order streams, high-order streams were found to have a significantly higher number of species ($F_{1,27} = 7.963$; $p = 0.009$) and total number of fish ($F_{1,27} = 12.96$; $p = 0.001$). We observed some potential threats to fish fauna in upper Appomattox River during our surveys including water pollution, siltation, stream bank erosion, and disturbance to riparian habitats.

SPATIAL AND TEMPORAL PATTERNS OF VEHICLE COLLISION-INDUCED WILDLIFE MORTALITY IN CENTRAL VIRGINIA. E. Robertson, J. Gibson, G. Cole, & S. Henkanaththegedara, Department of Biological & Environmental Sciences, Longwood University, Farmville, VA 23909. Roads could pose significant ecological impacts on environment and wildlife communities. Wildlife mortality due to vehicle collisions is one of the leading causes for population declines, population fragmentation and potentially local extirpations. The impacts of vehicle collisions on wildlife mortality in Virginia are poorly documented. We assessed the influence of road type (2-lane and 1-lane), seasons (spring and fall) and adjacent habitats (forest, agriculture and developed) on wildlife mortality rate. Highest average mortality rates were reported for gray squirrel (32.6%) followed by Virginia opossum (22.8%) and northern raccoon (13.0%). Although we found more wildlife mortality on 1-lane highway (59 total) compared to 2-lane highway (45 total), we failed to detect a significant road type effect. Similarly, we observed more mortality in fall (60) compared to spring (44), but failed to detect any seasonal effects. Additionally, there was more wildlife mortality reported in road segments adjacent to forests (56) followed by agriculture (49) and developed (11). This study generated the first data on wildlife mortality due to vehicle collisions for Virginia, and perhaps represents one of a few studies for the mid-Atlantic region. Understanding ecological implications of collision-induced wildlife mortality is important in implementing conservation measures.

BIODIVERSITY AND ECOLOGY OF TRIASSIC INSECTS OF VIRGINIA. Lucy E. Treado & Alexander K. Hastings, Virginia Museum of Natural History, Martinsville, VA 24112. Along the border of Virginia and North Carolina lies a fossil lagerstätte of Triassic flora and fauna known as the Solite Quarry. Some of the oldest known insect orders are found in the Cow Branch Formation shale bed at this site, many preserved with incredible detail of soft tissue. Less clear is the

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information regarding the aquatic environment in which they lived. Using a sample of 7,438 fossil insects and insect fragments that have been photographed and cataloged as part of a four-year digitization project, we attempted to address questions regarding environmental indicators. Based on our findings, this site was shallow, near-shore, and likely alkaline, due to the diversity and general size of the fossilized remains. The level of preservation indicated anoxia was likely, but cannot be definitively decided. (Supported by: The National Science Foundation iDigBio Fossil Insect Collaborative Thematic Collections Network Grant #1305066).

DIVERSITY, DISTRIBUTION AND ECOLOGY OF FRESHWATER CRAYFISH IN THE UPPER APPOMATTOX RIVER BASIN, CENTRAL VIRGINIA. D. Conner, C. Perry, & S. Henkanaththegedara, Department of Biological & Environmental Sciences, Longwood University, Farmville, VA 23909. Crayfish are virtually everywhere in the Southeastern United States, and represent the largest biomass in most aquatic systems. Although ubiquitous, very little is known about crayfish diversity, ecology and life-history, particularly in Central Virginia. We surveyed headwaters of the Appomattox River basin to document the diversity, distribution and ecology of crayfish. We documented 7 crayfish species belonging to 3 genera, including an invasive species. *Cambarus* sp. C is a widely distributed generalist that occupies all stream habitats and represents the most abundant species. *Cambarus longulus* is a rocky bottom specialist, and *C. diogenes* and *Fallicambarus uhleri* are primary burrowers. We also discovered 4 new populations of invasive *Procambarus clarkii* from Prince Edward County, which is a new Virginia state record. This exemplifies the diversity of crayfish in Central Virginia and shows the need for more detailed surveys to completely understand the diversity. State authorities will be able to use this information to update their records and implement more effective conservation practices.

Psychology

DO EMPATHETIC PEOPLE HAVE BETTER RELATIONSHIPS? A STUDY OF EMPATHETIC RESPONSES TO EMOTIONAL VIDEO STIMULI AND PERCEIVED SOCIAL SUPPORT. Nicole H. Wood & Antonina S. Farmer, Department of Psychology, Randolph-Macon College, Ashland, VA 23005. Empathy—the ability to relate to and match another's emotions—has been found in previous research to play a vital role in determining positive personal relationships. In this study, the researchers examined the association between social support and empathy, looking into how perceptions of social networks relate to both trait and state empathy. We hypothesized that there would be a positive correlation between these variables. Our sample consisted of 53 participants (26 male, 27 female) ranging from ages 18 to 50. Each participant completed multiple global self-report questionnaires (gauging trait empathy and social support) along with a video task in which a series of six video clips were chosen to elicit various emotional responses. Participants' facial expressions and emotional experiences (state empathy) were measured for each of the videos. Overall, the more empathetic participants generally felt (trait), the more empathetic they were to the video stimuli (state). Additionally, the greater the participants' perception of social support, the greater their emotional reactions (state empathy) were to the specific videos, but the participants' social support did not correlate with their trait empathy levels. Our findings add to existing research on the role of empathy in social relationships and suggest that people who experience greater social support do not necessarily see themselves as more empathetic overall but do respond more empathetically to specific situations. (Supported by: Shapiro Undergraduate Research Fellowship and William Craigie Research Grant).

IS IGNORANCE BLISS? THE RELATIONSHIP BETWEEN NEWS EXPOSURE AND ANXIETY LEVELS IN ADULTS. <u>Rebecca Reidy</u> & Antonina S. Farmer, Department of Psychology, Randolph-Macon College, Ashland, VA, 23001. While a substantial body of research has examined the relationship between stress and mental health, news exposure has yet to be studied as a daily hassle that could contribute to anxiety. The aims of this study are to 1) establish if there is a relationship between news exposure and anxiety levels, 2) evaluate whether anxiety levels change over time in relation to news exposure, and 3) establish if emotion regulation moderates these relationships. We recruited 502 participants (320 females, 181 males, and 1 intersex; M_{age} = 37.25 ± 12.93) to report on their exposure to current events, psychological symptoms (anxiety, depression, and stress), intolerance of uncertainty, experiential avoidance, and perseverative thinking over three waves. News related measures did not significantly change over time. Controlling for age, we did not find a relationship between news exposure and anxiety levels concurrently or prospectively. People with the greatest stress levels were those with high news exposure but a higher avoidance of the news. Lastly, participants who experienced greater enjoyment from the news tended to be less stressed and have greater psychological flexibility; moreover, prospectively, they were less likely to be depressed, when controlling for initial levels. In contrast, greater news exposure and less news enjoyment predicted more intolerance of uncertainty prospectively. Although our findings did not support our initial hypothesis about anxiety, news exposure and reactions to news predicted psychological distress and indicated a need for further, more nuanced research on this potential chronic stressor. (Supported by: Psi Chi Undergraduate Research Grant).

EVALUATING THE INFLUENCE OF MONEY ON CHILDREN'S CONCERN FOR OTHERS. <u>Tiffany T. Hwang</u>[1], Vikram K. Jaswal[1] & Charles W. Kalish[2], [1]Dept. of Psychology, University of Virginia, Charlottesville, VA 22904 and [2]Dept. of Psychology, University of Wisconsin–Madison, Madison, WI 53706. From infancy, children are motivated to acquire desirable resources, and from an economically rational perspective, accumulating resources should always be the goal. However, a growing body of research suggests that also from infancy, children prefer affiliating with members of their own group. We reasoned that when children make decisions, negotiating conflicts between material and social goals would be influenced by context. In Study 1, 6-year-olds chose between receiving a gift of a shiny sticker from Character A or a boring sticker from Character B. In a control condition (n = 24), 92% of children selected Character A's shiny sticker offer. In a gift condition (n = 24), Character B was said to be the children's teammate whereas Character A was said to be on the opposing team. This reduced the number of children selecting the shiny sticker to 33%, significantly fewer than in the control condition (Fisher's exact, $p < .001$). Children paid a price in order to affiliate with their teammate. To investigate whether children's priorities change when the context involves exchange, children in a nickel condition (n = 24) were given a nickel to exchange for a boring sticker from Character B (their teammate) or a shiny sticker from Character A. Results showed 63% preferred the shiny sticker from the opposing team member (Character A). In short, children's interest in acquiring desirable resources is surprisingly changeable: Affiliating with a teammate can be prioritized over acquiring material resources, but at least for 6-year-olds, this can be reversed when an exchange context is introduced.

DRUG DISCRIMINATION IN BEHAVIORAL STUDIES WITH RELATIONS TO BIOPSYCHOLOGY. <u>Amber N. Baldwin</u>[2], Tyler J. Braxton[1] & Joseph H. Porter[1], [1]Virginia

Commonwealth University, Richmond VA 23298 and [2]Thomas Nelson Community College, Hampton VA 23666. Drug Discrimination studies (DDs) emerged from Drug-induced state-dependent learning (SDL) in the 1950s. Today, Drug Discrimination is a behavioral study used to test stimulus effects with their corresponding receptor mechanisms. The DD procedure is an assay of effects of drugs in nonhuman to human animals. Several of these studies are conducted by a two-lever operant conditioning procedure. The training drug is given to a subject and the subject is then required to produce a response within a fixed-ratio. An appropriate response leads to reinforcement, while an incorrect response is not reinforced. Common goals of such studies offer a possible prediction of an agent in humans along with classifying and characterizing pharmacological effects/side effects. The Drug Discrimination procedure more generally holds unique characteristics in both neuroscience and behavioral pharmacology.

THE EFFECTS OF COMPUTER MEDIATED COMMUNICATION ON THE IMPLICIT BIAS TOWARD GAY MEN. JoEllen J. Blass & Cheryl L. Dickter, Dept. of Psychology, College of William and Mary, Williamsburg VA 23185. Sexual minorities experience discrimination on a regular basis which can lead to negative psychological, emotional, and cognitive consequences. Studies suggest that biases against minority group members can be reduced through intergroup contact. Recent research has used computer-mediated communication (CMC) in which participants think they are interacting online with an outgroup individual but are receiving a set of pre-determined responses. Positive CMC interactions have been demonstrated to reduce implicit bias towards racial and ethnic minorities. The goal of this study was to examine the effects of CMC on attentional bias against sexual minorities. Attentional bias is thought to reflect the automatic encoding and orienting of social categorization, which could have implications for prejudicial behavior. Heterosexual participants ($N = 171$) communicated online with a male identified as either gay or straight who exhibited stereotype consistent or inconsistent behaviors. Participants then completed a Dot Probe task assessing their implicit attention to images of heterosexual and homosexual couples. Results revealed that participants interacting with gay males who displayed stereotype-inconsistent behaviors showed implicit attentional bias away from homosexual couples; no other groups differed significantly from zero. These findings suggest that CMC may lead to a reduction of attentional bias towards sexual minorities. Because CMC is simple to implement, this could be used to reduce prejudice in everyday settings including school and work.

DOES LANGUAGE TRANSLATE TO EXECUTIVE FUNCTIONS? INVESTIGATING THE BILINGUAL ADVANTAGE IN EXECUTIVE CONTROL. Melina L. Knabe & Sarah N. Blythe. Dept. of Biol., Washington and Lee Univ., Lexington VA 24450. Given that twenty-percent of the U.S. population speaks a language other than English at home, it is imperative to assess the effect of a second language on brain structure and function. The *bilingual advantage hypothesis* claims that command over two languages leads to enhanced non-linguistic cognition. The need of a bilingual to maintain both languages active simultaneously, inhibit one, and flexibly switch between both may transfer to executive control (EC) functions. The present study investigates the effect of mono- and bilingualism on inhibitory control and cognitive flexibility across age groups. Implementing an online format, mono- and bilinguals (ages 18-89, $n = 334$) of diverse language parities completed a language background and demographic questionnaire. EC performance was assessed using a Simon task, task-switching paradigm, and directed forgetting (DF) paradigm. In addition, Shipley-II vocabulary and block patterns tests served to assess crystallized and fluid intelligence. It was hypothesized that bilinguals would outperform monolinguals on all tasks and

that this difference would be most pronounced in older ages. Further, it was hypothesized that amongst bilinguals, age of active onset, language proficiency, and amount of language switching would be most predictive of outcomes. A moderated regression showed that after controlling for various lifestyle factors, middle-aged bilinguals showed faster reaction times than their monolingual counterparts, suggestive of a Bilingual Executive Processing Advantage (BEPA) rather than a Bilingual Inhibitory Control Advantage (BICA). Results also revealed that this trend disappeared in the task-switching paradigm, a more demanding task. Finally, young bilinguals showed greater item recall on the DF paradigm, suggesting improved maintenance of information in the presence of interfering information. Multiple hierarchical regressions revealed no significant predictive value of various bilingual experiences on task outcomes, except for a putative role of age of active onset on Simon effect outcomes. (Supported by: The Virginia Academy of Science Undergraduate Research Grant).

Posters

COMPULSORY EQUALITY: CHILDREN THINK RESOURCES MUST BE DIVIDED EQUALLY. Marissa B. Drell & Vikram K. Jaswal, Dept. of Psychology, Univ. of Virginia, Charlottesville, VA., 22904. How limited resources should be distributed is one of the most vexing problems in society. While an equal distribution may be ideal, there are circumstances under which inequality can be acceptable. Here, we asked whether participants would evaluate inequality as permissible if someone chooses to take less than her fair share. In Study 1, 6-year-olds, 8-year-olds, and undergraduates watched a series of cartoons involving two characters and four pieces of candy. One character expressed a desire for only one piece and took just one, and the other character took the remaining three. Participants decided whether the distribution was "okay" or "not okay." Most undergraduates said this type of inequality was "okay," but most 6-year-olds and half of the 8-year-olds said it was "not okay." In Study 2, 6-year-olds saw the same scenarios except that after taking just one of the four available candies, the first character gave permission for the other character to take the remainder and actually gave them to her. Surprisingly, most 6-year-olds continued to indicate that the unequal distribution was "not okay." Together, the results from these two studies suggest that 6-year-olds have an extremely strong aversion to unequal outcomes. They only attend to the fact that one character possesses more items than the other and disregard other factors, including desire, choice, permission, and generosity.

DAGL-A KNOCKOUT MICE DISPLAY A PROFOUND IMPAIRMENT IN SPATIAL LEARNING AND MEMORY. Lesley D. Schurman[1], Moriah C. Carper[1], Daisuke Ogasawara[2], Benjamin F. Cravatt[2], & Aron H. Lichtman[1], [1]Virginia Commonwealth University, Richmond VA, 23284, and [2]Scripps Research Institute, La Jolla CA, 92037. Diacylglycerol lipase (DAGL-α), which is highly expressed on neurons, represents a primary biosynthetic enzyme of the endocannabinoid 2-AG. DAGL-$\alpha^{-/-}$ mice show ~80% reduction in brain 2-AG, as well as impaired endocannabinoid-regulated synaptic plasticity, and compromised hippocampal neurogenesis. Here, we examine whether genetic deletion or pharmacological inhibition of this enzyme impairs performance in the mouse fixed platform Morris water maze (MWM) task. DAGL-$\alpha^{-/-}$ mice failed to acquire MWM reference memory and showed no evidence of spatial memory expression; however, showed good performance in a cued MWM task (i.e., in which the platform was made visible), ruling out sensorimotor or motivational deficits. The DAGL–α inhibitor DO34 produced small, yet significant dose-dependent delays in MWM acquisition rate, without any performance

deficits in either memory expression or an extinction task. The severely impaired MWM performance of DAGL-$\alpha^{-/-}$ mice compared to DO-34-treated mice may reflect developmental consequences of constitutive DAGL-α gene deletion. The impaired acquisition rate, but not expression or extinction of DO34-treated mice may suggest that DAGL-α contributes to the homeostatic regulation of specific aspects of spatial learning and memory. These findings point to the regulatory, and perhaps developmental, importance of this 2-AG biosynthetic enzyme in spatial learning and memory. Supported by: NIH NRSA award IF31NS095628-01A1 and VCU School of Pharmacy start-up funds.

Structural Biology, Biochemistry and Biophysics

Not yet submitted